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Biology of *Caryedon crineus* Arora (Coleoptera: Bruchidae), a seed pest of *Cassia occidentalis* (L.)

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ABSTRACT: Biology and behaviour of *Caryedon crineus* Arora, infesting pods and seeds of *Cassia occidentalis* (L.) was studied. Female beetle lays 16–27 eggs on pods and seeds of host plant during nights, which hatch after about 9 days. All larval instars are voracious feeders and their development is completed inside the host seed. Larval duration lasts for 23–27 days and last instar larva emerges out of seed by cutting a hole and then undergoes pupation outside. Pupal duration inside cocoon lasts for 51–60 days during favourable period and 191–220 days during adverse conditions and adult emerged out after 85–96 days. © 2007 Association for Advancement of Entomology

KEYWORDS: *Caryedon crineus*, biology, *Cassia occidentalis*

INTRODUCTION

Caryedon crineus Arora (Bruchidae: Coleoptera) is a pest of *Cassia occidentalis* (L.) (Panwar) in India (Arora, 1977). Adults appear in the field in the month of April after passing the winter as hibernated pupa. Biology and ecology of species of *Caryedon* have been studied by various workers. Pajni and Mann (1979); Pierre and Huignard (1990) studied the biology of *Caryedon serratus* (Oliver). Conway (1983) showed that mating, oviposition and development in *C. serratus* occurred in considerable depth in jute bags but fourth instar pupates outside the bag stacks. Halle *et al.* (2002) studied the comparative biology of tamarind beetle, *C. serratus* on nine different hosts in laboratory and Thakur and Banyal (2003) demonstrated the life history of *C. crineus*. Southgate (1979) observed three different sites of cocoon formation among some Nigerian species of genus *Caryedon*. The taxonomic position of *C. crineus* was reported by Arora (1977, 1978). Except for life history and larval parasitoid (Thakur and Banyal, 2003, 2005) there is no information on biological aspects of this species. Hence studies were made on the biology and behaviour of *C. crineus*.

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MATERIALS AND METHODS

Adults of *C. crineus* emerged in the month of April were kept in pairs (one male and one female) along with seeds and pods of *C. occidentalis* in wire mesh cages of $10 \times 12 \times 12 \text{ cm}^3$ and 500 ml glass jars with their open end covered with muslin cloth, in the laboratory. Observations were made on behaviour of adults and immature stages. The number of eggs laid per female, their shape and size and incubation period were also observed. Morphometric studies of eggs and first instar larvae were carried out. Eggs of almost equal age were marked and observations were made daily. The larval instars duration, pupal duration, time taken for completion of life cycle, longevity of insects and number of generations completed in a year were recorded.

RESULTS

Behaviour

The male *Caryedon crineus* Arora was smaller in size than the female and had vertical pygidium. Adults of both the sexes showed brisk activity during the evening and early morning hours and remained hidden and inactive during daytime. Adults feigned death when touched or disturbed, turned upside down, retracted neck, pulled their legs and antennae close to the body and remained motionless but regained normal posture and activities after 10–20 seconds. Mating occurred inside the pods among the seeds during night time because neither mating nor egg laying has been observed during day time.

Mating

Male recognized the female insect from a close distance, chased her and continuously tapped her back or pygidium with its antennae. Male then put its antennae on the back of the female and the female remained motionless. Male insect then mounted on the back of the female and held her abdomen with his fore and mid legs. During the initial time of mating, the female pushed the male off her back with her hind legs, but persistent attempts of the male facilitated the insertion of already extruded aedeagus into the genital opening of the female resulting in successful copulation. During copulation female remained stationary and jerking movement of the male insect was observed. When another male tried to dislodge the copulating couple, the female moved ahead and pulled the inseminating male on her back.

After 10–20 min of copulation the female pushed the male away with her hind legs. After copulation male and female cleaned their mouthparts and antennae with fore and mid legs and genitalia or ovipositor with hind legs and then started moving away. When two males tried to mount the same female, the stronger one succeeded in the attempt and weaker moved away in search of another receptive female.

Oviposition

Gravid female of *C. crineus* started laying eggs 7.1 ± 1.1 days after emergence and eggs were laid during night time only. Females of *C. crineus* followed one seed -

one egg pattern but pods of the host plant contained 1–8 eggs. Females of *C. crineus* after laying their full quota of eggs became sluggish and died 3.3 ± 1.3 days after the oviposition was completed.

Before depositing an egg, the female examined the seed carefully with her antennae. It even turned over the seeds by thrusting her head underneath the seeds and pushed them. After searching a seed, the female lowered her abdomen, extruded the ovipositor, rubbed it on the surface of the seed and released a sticky fluid for attachment of the egg. The egg was then deposited on the treated surface with its broader end coming out first from the ovipositor by shaking abdomen. The whole process of depositing an egg on the surface of seed or pod took about 2–3 min. Oviposition period of a female of *C. crineus* extended for 9.0 ± 2.2 days and maximum of 9 eggs were deposited on the 2nd night of her oviposition period.

A single female of *C. crineus* laid 21.8 ± 3.2 eggs. Freshly laid eggs were rusty or light orange, oval in shape and measured 0.84 ± 0.06 mm in length and 0.496 ± 0.03 mm in width. Eggs laid in the beginning and near the end of oviposition period were empty and without ooplasm and failed to hatch. In the absence of seeds egg laying was postponed and in the prolonged absence of the seeds eggs were deposited on the walls of the test tube or petridish. Such eggs on the walls of culture vessels produced normal first instar larvae inside the egg shell but the larvae did not hatch out and died inside the egg shell within 2–3 days.

Incubation period lasted for 8.9 ± 0.37 days under natural atmospheric conditions (24°C – 39°C temperature and 53%–98% relative humidity). Egg shell became transparent before hatching and developing first instar larvae were visible. But this visibility was lost before hatching due to deposition of the larval frass inside the egg shells.

Larval development

First instar larvae hatched out from the ventral surface of the eggs, penetrated vertically into the host seeds and then turned at an angle to feed on the contents of the host seeds. On the way into the host seed first instar larva consumed the contents of seed and whitish and light orange frass was deposited inside the empty egg shell. The entrance hole into the seed was closed by the first instar larva with its faecal matter but could be identified as a white mark on the seed.

First instar larvae measured 0.469 ± 0.02 mm long and 0.237 ± 0.01 mm broad, each having large brownish rounded head and elongated tapering body covered with numerous white shining setae of variable size. The body of larva was divided into head, thorax and abdomen. Head possessed the necessary appendages, viz. clypeus, labrum, labium, a pair of mandibles, a pair of maxillae and a pair of antennae. Thorax possessed three pairs of legs and abdomen was tapering posteriorly and devoid of any appendages. First instar larva possessed H-shaped prothoracic plate with six pairs of dark, highly sclerotized teeth at its outer margin. The prothoracic plate facilitated the entry of larva into the host seed. Second and successive instar larvae did not possess the prothoracic plate. Each larval instar exuded a yellowish fluid to soften the contents of the seed. Head was smaller but deeply retracted in prothorax in successive instar

larvae. All the larval instars were voracious feeders and kept on feeding all the time except during moulting and consumed the seed contents completely leaving behind seed coat only.

Further development of first and successive instar larvae was completed inside the host seed. Larvae did not shift from one seed to another even when the contents of the initially infested seed were exhausted probably due to the inability of second and successive instars to possess the chiseling apparatus, the H-shaped prothoracic plate. Last instar larva came out of the seed 34.1 ± 1.59 days after oviposition (Table 1). It wandered among the seeds and pods in search of suitable place for cocoon formation. This stage lasted for 4–6 h only. Some last instar larvae were attacked by a parasitoid, *Eurytoma raoi* Narendra (Hymenoptera: Eurytomidae) and killed during development inside the cocoon. Cocoon was formed either outside a pod or inside the pod among the seeds.

Occlusion

Adult insect emerged out after cutting an incomplete circular lid at one end of cocoon and a complete circular window in the pods of the host plant. The pupae in the cocoons formed in October did not produce adult insects immediately but overwintered as pupal instar till next April. Therefore, pupal duration inside the cocoon lasted for 55.8 ± 3.01 days during favourable period (April to September) and 205.4 ± 8.82 days during unfavourable period (October to March). Life cycle of *C. crineus* was completed in 89.9 ± 3.44 days and total longevity of the insect (including egg, larval, pupal and adult stages) lasted 109.3 ± 3.71 days under natural atmospheric conditions (24°C – 39°C temperature and 54%–98% relative humidity) during the favourable period.

DISCUSSION

In *Caryedon crineus* mating and egg laying occurred during night time similar to *C. serratus* (Pajni and Mann, 1979). Behaviour of gravid female to deposit single egg on healthy and unwrinkled seed and pod of host plant is comparable with *Callosobruchus maculatus* (Huignard *et al.*, 1985; Wasserman, 1985); *C. theobromae* (Thakur and Banyal, 2004); and some other species of genus *Caryedon* (Southgate, 1979).

In the absence of seeds and pods of host plant the females of *C. crineus* delayed oviposition for 3–4 days and finally laid eggs on the walls of the test tubes and petridishes but such eggs failed to hatch out similar to *Zabrotes subfasciatus* (Pajni and Jabbal, 1986). Eggs laid by *C. crineus* towards the end of oviposition period were without ooplasm like the eggs of *Callosobruchus chinensis* where size and number of eggs decreased with maternal age (Yanagi and Miyatake, 2002). Mating duration of 10–20 min and 21.8 ± 3.2 eggs laid per female in *C. crineus* are comparable with 25–40 min long mating and 26 eggs per female in *C. serratus* (Pajni and Mann, 1979).

First instar larvae of *C. crineus* hatched out from the ventral surface of the eggs and penetrated into host seed or pod with the help of a H-shaped prothoracic plate

TABLE I. Duration and dimensions of different stages observed during the life cycle of *Caryedon crineus* (mean \pm standard deviation).

Incubation period (days)	Egg dimensions (mm)		Dimensions of first instar larvae (mm)		Egg to pre-pupal duration (days)	Pupal duration during favourable period (days)	Pupal duration during unfavourable period (days)
	Length	Breadth	Length	Breadth			
8.9 \pm 0.37	0.840 \pm 0.06	0.496 \pm 0.03	0.469 \pm 0.02	0.237 \pm 0.01	34.1 \pm 1.59	55.8 \pm 3.01	205.4 \pm 8.82

like in *C. serratus* (Pajni and Mann, 1979), *C. theobromae* (Thakur and Banyal, 2004) and *Bruchus pisorum* (Pajni and Sood, 1984). Exhibiting similarity to the larvae of *Bruchidius atrolineatus* and *C. maculatus* (Germain *et al.*, 1987) larvae of *C. crineus* do not shift from one seed to another even in the insufficiency of food contents in previously infested seed. Last instar larva of the insect under study formed cocoon outside the infested seed among the debris like *C. serratus* (Conway, 1983).

C. crineus completed its life cycle in 89.9 ± 3.4 days on *Cassia occidentalis* seeds whereas life cycle of *C. serratus* was completed in 51 days on tamarind and in 91.63 days on *Bauhinia rufescens* seeds (Halle *et al.*, 2002). Adults of *C. crineus* appeared in the field in the month of April after passing the winter as hibernated pupa similar to *B. pisorum* (Pajni and Sood, 1976). Pupal duration of *C. crineus* inside cocoon lasted for 205.4 ± 8.82 days during unfavourable period. A similar prolonged life cycle of 6–8 months during cooler months has been observed in *C. chinensis* (Yoshida *et al.*, 1986).

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Ecdysone-neurosecretion interaction during vitellogenesis in *Poecilocerus Pictus* Fabr. (Orthoptera: Acrididae)

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ABSTRACT: Different concentrations of β -ecdysterone (No-E-2003) in solution in the newly emerged female insect from day 1 to 30 days during the genotrophic cycle stimulates vitellogenin synthesis in the adipose tissue. This was found to be the effect of 'A' type of neurosecretory cells and inhibition of its hormone secretion regressed the activity of *Corpus allatum* (CA). On the contrary, the 'B' type cells of the *pars- intercerebralis* of EDNH system remain unaffected and their secretions reach the hemolymph via *Corpus cardiacum* (cc) and acts in conjunction with ecdysone to regulate the uptake of vitellogenin in the ovaries. This uptake is rather slower in the presence of juvenile hormone (JH). It is concluded that 'B' type neurosecretory cell hormone is comparable to a parsian -Lom OMP described by Girardie *et al.* (1996a,b) in *Locusta migratoria* that influence egg development.

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INTRODUCTION

A lot of research has been done on the role of neurosecretion and juvenile hormone (JH) on insect ovarian development (Johannsson, 1955; Nayar, 1955; Wigglesworth, 1936, 1985; Highnam, 1962a; Dogra and Tandon, 1964; Saini, 1966, 1971; Dogra and Ewen, 1970; Gilbert, 1985; Girardie *et al.*, 1996a,b). JH plays a central role in egg production (Wigglesworth, 1985; Strong, 1965a) and acts on the fat bodies to initiate vitellogenin synthesis. The regulation of ovarian development via JH has been fully elucidated by many authors (Engelmann, 1983; Gilbert, 1985) but little work has been done on hormonal homeostasis in female reproduction.

Horn (1971); Lagueux *et al.* (1977) have suggested that the ecdysteroids in the adult female might play a role in reproduction, while according to Stay and Tobe (1981) the secretion of ecdysteroids from the late stage ovaries inhibit JH synthesis.

It is also well established that fat bodies synthesized vitellogenin and the synthesis is activated in the presence of 20-hydroxyecdysone (Bohm *et al.*, 1978). Injection of micro-quantities of 20 hydroxyecdysone stimulated vitellogenin synthesis in the mosquito *Aedes aegypti* (Fallon *et al.*, 1974). A factor like egg development

neurosecretory hormone (EDNH) was found to stimulate the synthesis of both ecdysone and vitellogenin (Lea, 1972, 1982). According to (Rossignol *et al.*, 1981) oversecretion of ecdysone affected JH secretion, hence the ecdysteroids may be inhibiting CA (Fallon *et al.*, 1974; Stay and Tobe, 1981). The role of ecdysone in adult insects is rather obscure, especially when its level is low. After noticing these ambiguities, in the present studies an attempt has been made to elucidate the effect of ecdysone on the neurosecretory system including CA along with its role on vitellogenin synthesis from adipose tissue and ovarian development in *P. pictus*.

MATERIALS AND METHODS

For the present study *Poecilocerus pictus* Fabr. (Orthoptera, Acrididae) has been used. The insects were collected from *Calotropis* plant during May/June to October and fresh *Calotropis* leaves were provided daily *ad libitum*. The sixth instar nymphs took about 6 to 8 days to become adult, if fed properly.

The newly emerged adult insects were sorted out and they were injected with different concentrations of β -ecdysone from day one to thirty when they start egg laying. Various concentrations of ecdysone was injected after every 24 hrs. The tissues studied are neuroendocrine complex, ovary and adipose tissue.

The concentration of hormone used were 1 μ g, 7.5 μ g, and 15 μ g per ml of acetone. 2 μ l of each concentration (containing 2 μ g, 15 μ g, 30 μ g ecdysone each) was injected to each experimental insect of different age group from day one to 3 days, 8 days, 15 days, 20 days, 25 days, and 30 days, by Himilton Micro-syringe. The same concentration of acetone without ecdysone was injected in to each control insect.

The neuroendocrine complex including the neurosecretory cell groups, *Corpus cardiacum* and *Corpus allatum* were examined. The adipose tissue and ovaries were also examined during growth, reproductive period and during maturation period and progress of vitellogenesis relating to endocardial activity was also co-related.

The neurosecretory cells of the pars-intercerebralis (both of A and B type NS cells) were examined with different staining methods like, Paraldehyde-fuschin (Gomori, 1950; Halmi, 1952; Gabe, 1953) referred as PF, and Chromo Haematoxylin – Phloxine (Gomori, 1950) referred as (CHP). For making observations on A-cells, technique of Ewen (1962) and Delphin (1965) were followed. Alcian blue after permanganate oxidation, counter stained with Phloxine followed by phosphotungstic acid was followed to stain different types of neurosecretory cells. Performic acid–alcian blue technique for cystine comparing NS also was used (Adams and Sloper, 1956).

Hematoxylin–eosin staining was performed to examine the adipose tissue and ovaries. For nucleic acids, the methyl green pyronine method Bonhag (1955) (Baker and Barbara, 1955; Pearse Everson, 1960) was applied. For staining proteins methods of and Pearse Everson (1960) were used.

RESULTS

Neurosecretory Cells: Four different types of neurosecretory cells A, B, C and D types in the median group and about 8–10 lateral cells located in the lateral area

of brain could be observed. Cell types could be identified by the different staining techniques used. A and B cells showed marked secretory activity and the secretion could be seen even in their axons while in the C and D type cells no secretory activity could be noticed; but vacuoles were noticed in these cells. Further there was no neurosecretory material noticed in the axons, and hence their activity could not be correlated with reproduction.

About 80–100 different NS cells could be localized in the median group. The A-type cell could be identified by PF technique and there were about 80 cells showing positive reaction to NS. Purple inclusions were noticed by PF and Alcian blue azan technique as seen by (Ewen, 1962; Delphin, 1965). These cells showed cyclic activity and they can play a major role in moulting and reproduction. The neurosecretion reached corpus cardiacum, and some neurosecretory axons reached the corpus allatum also. B-type cells showed faintly reddish neurosecretory material and their axons terminate inside the *Corpus cardiacum* (CC). The secretory activity of A-and B-type cells showed a correlated activity with vitellogenin synthesis, ovarian development, maturation and vitellogenesis, and the correlated NS activity has been depleted in (Fig. Plates 1 and 2 Fig. 1–5).

Different concentrations of ecdysone showed marked secretory changes in all types of cells and also during vitellogenin synthesis, ovarian development, yolk synthesis and vitellogenesis.

The ovarian development in normal female

In newly emerged female, vitellogenesis progresses and egg laying takes place within 28–30 days. Vitellogenesis starts from 15 days after emergence that is being completed with in about 25 days.

In three-day-old female, the median cells of A-type have some scattered granules that indicate secretory activity. There is a lot of secretory material in the axons but only little material is noticed inside the cell. The cells of corpus allatum are closely packed but neurosecretory material is noticed in the axons reaching the *Corpus allatum*.

Within 5–8 days, most of the neurosecretory material is passed to the CC. Cells of the *Corpus allatum* are still closely packed but start increasing in size. Within 13–15 days of emergence, the neurosecretory material was found to move from the body of the cell to the CC. But by this time yolk synthesis has started and at the same time the CA is found enlarged.

Within 19–20 days of emergence blue black granules are noticed in the body of A-type cells and the *Corpus allatum* and found enlarged. In the ovary, the terminal Oocyte has grown to its full size.

After 25 days from emergence, neurosecretory (NS) material is found to be filled with in the cell but no material is noticed in the axons. Vitellogenesis was also completed during this time.

Within 29–30 days from emergence, A-type cells refilled with purple coloured material, but the axons are not prominent, size of the *Corpus allatum* found to be reduced. Within 25 to 30 days chorion formation is noticed and egg laying started.

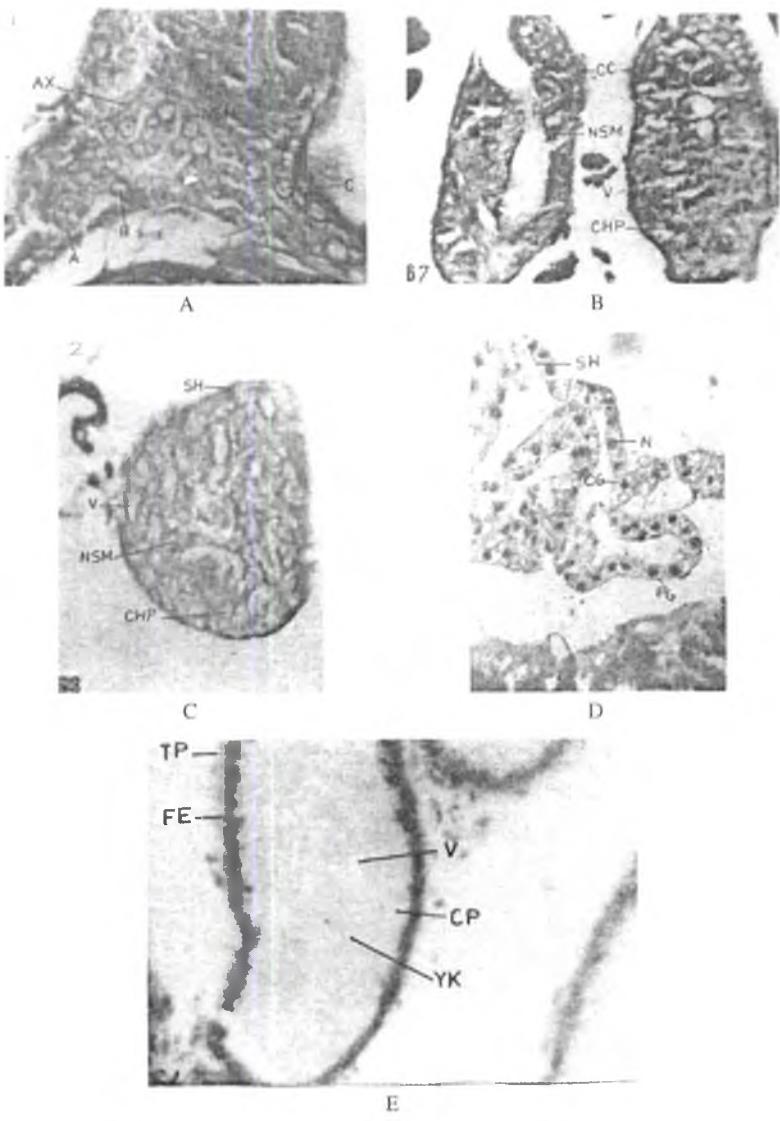


PLATE-1

FIGURE 1. A. Section of brain of adult *P. pictus* after 15 days treated with 0.03% Ecdysone ($\times 200$) PF staining showing A, B and C type of neurosecretory cells with NSM in it Ax-Axons. NSM=Neurosecretory material. B. Section of corpus cardiacum of adult *P. pictus* after 15 days treated with 0.03% Ecdysone ($\times 200$) P.F. with NSM in CHP-Chromophil cells. C. Section of corpus allatum of adult *P. pictus* after 15 days treated with 0.03% Ecdysone ($\times 200$) P.F. showing material stored in the posterior portion. SH-Sheath, V = Vacuoles, NSM = Neurosecretory material, CHP = Cells of corpus allatum. D. Section of adipose tissue of adult *P. pictus* after 15 days treated with 0.03% Ecdyson showing distinct nucleus and Vacuolated Cytoplasm ($\times 100$) M.G/P. E. Section of ovary of adult *P. pictus* after 15 days treated with 0.03% Ecdyson showing vacuoles in the ooplasm ($\times 200$) M.G/P.

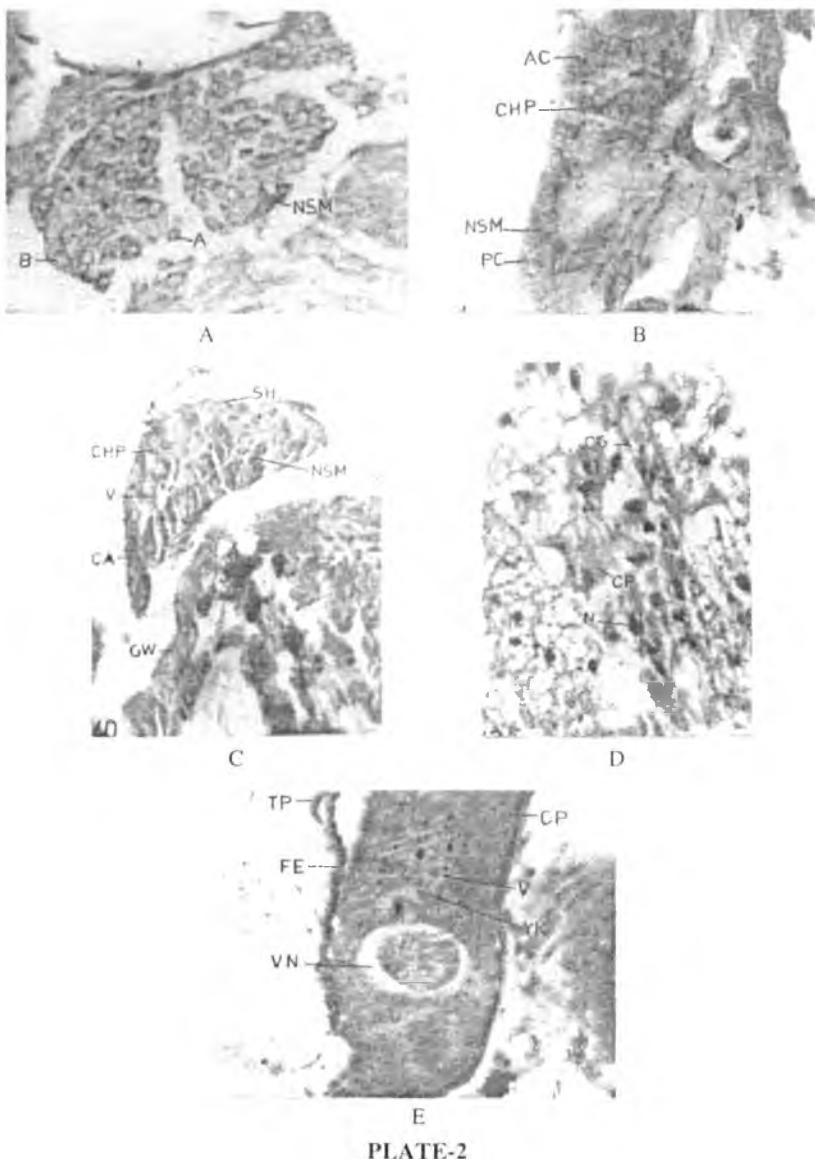


PLATE-2

FIGURE 2. A. Section of brain of adult *P. pictus* after 15 days treatment with 0.15% Ecdyson ($\times 200$) P.F showing A, B and C type of cells with NSM. B. Section of corpus cardiacum of adult *P. pictus* after 15 days treatment with 0.15% Ecdyson showing NSM in it ($\times 200$) P.F. C. Section of corpus allatum of adult *P. pictus* after 15 days treatment with 0.15% Ecdyson ($\times 200$) with cells of corpus allatum and NSM = Neurosecretory material in corpus allatum in cells. D. Section of adipose tissue of adult *P. pictus* after 15 days treatment with 0.15% Ecdysone ($\times 200$) M.G./P. E. Section of ovary of adult *P. pictus* after 15 days treatment with 0.15% Ecdyson ($\times 200$) M.G./P. FE = Follicular epithelia.

In control insects, major events are the same as that in the normal insects. But in the experimental insects which were injected with different concentrations of ecdysone, the synthesis of neurosecretory material in cells was rather slow and they were filled with materials as the transport also was slower and thus neurosecretory material reaching the *Corpus allatum* was inhibited and the corpus allatum was found regressed. There was no juvenile hormone secretion. Further, there was vitellogenin synthesis as usual and uptake of vitellogenin took place. But it was rather slower than that takes place in the presence of Juvenile hormone. On the contrary the B cells show normal secretion.

The neurosecretory activity showed that the secretion of B cells and ecdysone in conjunction play a role in the uptake of vitellogenin to the ovaries for yolk synthesis and vitellogenesis. This process is rather slower than that takes place in the presence of juvenile hormone. In comparison with normal vitellogenesis, this takes place between 25–30 days. Egg laying is also delayed and takes place after 30–35 days.

DISCUSSION

The insects were injected with ecdysone and with different concentrations it was found that the ecdysone plays different roles. It stimulated the synthesis of vitellogenin in adipose tissues showed increased RNA activity and vacuole formation. These observations are similar to that described by Fallon *et al.* (1974) and Hagedorn (1983) in mosquito. The injection of ecdysteroids to cockroaches reduced the activity of *Corpus allatum* (Friedel *et al.*, 1980; Lanzrein *et al.*, 1981, Rossignol *et al.*, 1981). Stay and Tobe (1981) observed that administration of ecdysteroids inhibit the activity of *Corpus allatum* and juvenile hormone synthesis. Present studies showed that different concentrations of ecdysone in *P. pictus* have an inhibitory effect on *Corpus allatum* which is mediated through prothoracico tropic hormone, from A-type of cells whose hormone reaches there by the axons to *Corpus allatum*. Although the activity of gonadotropic hormone (JH) was inhibited, vitellogenin synthesis in adipose tissue was found stimulated even though there was slow uptake of vitellogenin and yolk synthesis noticed in the ovaries in conjunction with some other hormone, the egg development hormone, EDNH Lea (1972). Ecdysone affects the secretion and transport of A-cell hormones of the EDNH complex while the B-cells and other cells of the EDNH complex are not affected and their hormone reaches the hemolymph via the *Corpus cardiacum*. Ecdysone acts in conjunction with the hormone to help vitellogenin uptake.

Mechanism of vitellogenesis is slower in the presence of JH. Hence a delay in vitellogenesis is noticed. The maturation period and vitellogenesis take place with in 15 to 25 days in normal females while in the experimental females it takes 25 to 30 days (egg laying takes place in 35 days). The hormone of B-type NS cells which is unaffected by the ecdysone is similar to that of Lom OMP parsian described by Girardie *et al.* (1996a) in *Locusta migratoria*. In short ecdysone appears to be a raw hormonal material that acts in conjunction with other hormones showing different modifications required for the physiological requirement of the insect.

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Study of beetles (Coleoptera) associated with stored products in Kolkata, India

1. Genus *Carpophilus* Stephens (Nitidulidae)

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ABSTRACT: Six species of *Carpophilus* Stephens are reported from 18 different commodities. Of these, *Carpophilus freemani* Dobson, *C. obsoletus* Erichson, *C. pilosellus* Motschulsky, *C. delkeskampi* Hisamatsu and *C. marginellus* Motschulsky are new records from stored products in India. Diagnostic features, distribution and habitat records of all six species are dealt with and a key to these species of *Carpophilus* is given. © 2007 Association for Advancement of Entomology

KEYWORDS: Nitidulidae, *Carpophilus* Stephens, stored product pests

INTRODUCTION

Carpophilus Stephens is a large genus, comprising about 193 species (Jelinek, personal communication, 2006) that includes most of nitidulid species infesting stored products. This genus can be recognized by the following characters: head distinctly narrower than pronotum, clypeus slightly indistinct and margined by a depression on either side; eyes large; antennae little longer than head with compact, flattened, rounded or oval club; labrum bilobed; pronotum about as broad as elytra; scutellum broadly rounded posteriorly; elytra short, exposing two apical abdominal segments. Species of *Carpophilus* Stephens infesting stored products, especially in Europe, were dealt with earlier by Dobson (1954a,b, 1956, 1960). Grouvelle (1913) mentioned four species of *Carpophilus* from India, namely, *Carpophilus dimidiatus* (Fabricius), *C. hemipterus* (Linnaeus), *C. ligenus* Murray and *C. mutilatus* Erichson encountered commonly in stores. Aitken (1975) reported these four species intercepted from Indian products imported to U.K. and altogether nine species of *Carpophilus* from the world. In the present study, five more species namely, *C. freemani* Dobson, *C. marginellus* Motschulsky, *C. obsoletus* Erichson, *C. pilosellus* Motschulsky and *C. delkeskampi*

Hisamatsu, besides *C. hemipterus* (Linnaeus) were detected during the course of an intensive survey of stored product beetles from Metropolitan Kolkata during 1982–85. Systematic accounts of these species are given and a key to their identification is incorporated.

SYSTEMATIC ACCOUNT

Family Nitidulidae

Subfamily Carpophilinae

Genus *Carpophilus* Stephens

Carpophilus Stephens, 1830, *Ill. British Insects*, **3** 50.

Carpophilus delkeskampi Hisamatsu

Carpophilus hemipterus (nec. Linnaeus) Stebbing, 1914, *Indian Forest Insects, Coleoptera* 108.

Carpophilus delkeskampi Hisamatsu, 1963, *Entomon. Rev. Japan*, **15**: 61.

Diagnosis

Brownish yellow with paler legs, elytra yellowish with patches of brown near base, sparsely and finely pubescent (Fig. 1).

Size: 2.50–2.65 mm.

Material studied

8 exs. India: W. Bengal, Kolkata, Sovabazar, *ex. maize grain*.

19.iii.1982; P. K. Basak; 2 exs, Kolkata, Posta, *ex. cassia leaf*, 2.vii.1982; P. K. Basak; 3 exs. Howrah, Bandhaghat, *ex. cotton*, 17.11.1982, 1 ex, Kolkata, Chitpur, *ex. jute thread* 23.vi.1983, P. K. Basak. 2 exs.

Distribution

India: West Bengal (new record); Sri Lanka.

Remarks

There is no previous record of habitat of this species and in the present study it has been recorded from maize grain, cassia leaf, cotton and jute thread.

Carpophilus freemani Dobson

Carpophilus freemani Dobson, 1956. *Entomologist's mon. Mag.*, **92**: 41.

Diagnosis

Brownish black with paler elytra and moderately long and recumbent pubescence (Fig. 1(2)).

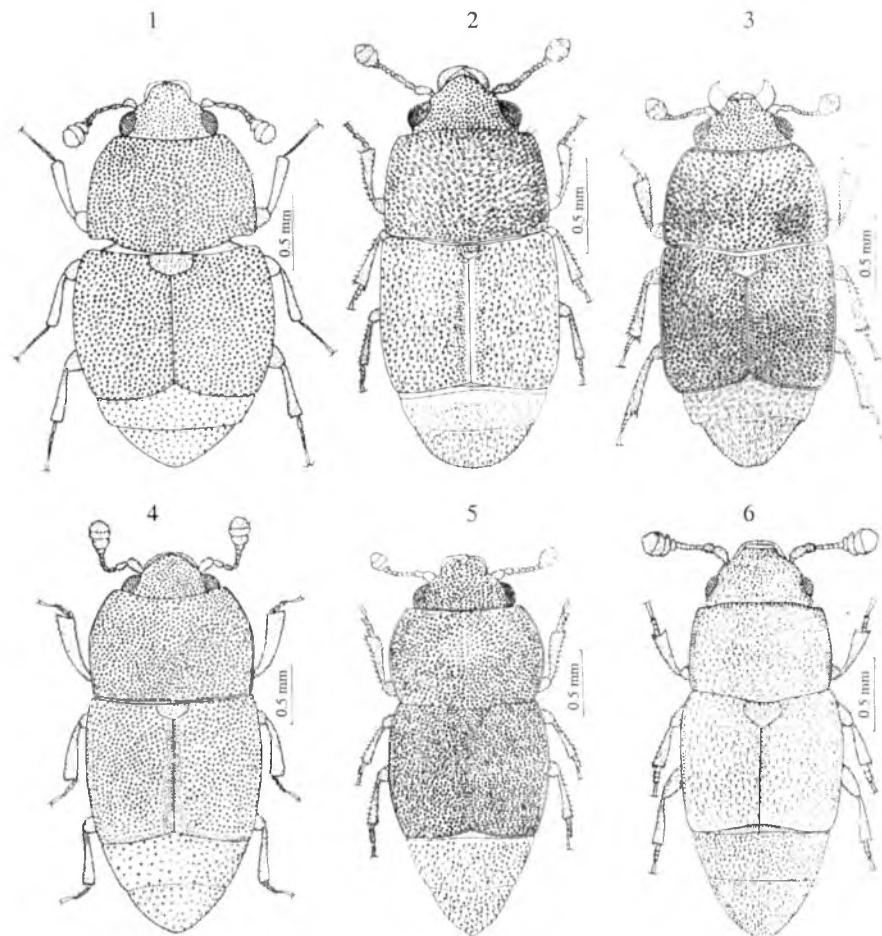


FIGURE 1. 1. *Carpophilus delkekampi* Hisamatsu, Dorsal view. 2. *Carpophilus freemani* Dobson, Dorsal view. 3. *Carpophilus hemipterus* (Linnaeus), Dorsal view. 4. *Carpophilus marginellus* Motschulsky, Dorsal view. 5. *Carpophilus obsoletus* Erichson, Dorsal view. 6. *Carpophilus pilosellus* Motschulsky, Dorsal view.

Size: 2–23 mm.

Material studied

9 exs. India. Howrah, Railway yard, ex. tamarind 17.ii.1982; P. K. Basak; 7 exs., Howrah, ex. fish baskets and gunny sacs, 15.i.1982; P. K. Basak; 2 exs.

Distribution

India: West Bengal; England; North America; South America.

Remarks

Dobson (1956) described this species from specimens collected from Brazil nuts. It also infested many kinds of moist decomposing vegetable matter, including apple and peach drops, corn ear refuse and weevil infested nuts (Connell, 1956; Aitken, 1975) recorded this species from Brazil nuts, maize, bones, and sheanuts. In the present study it was recorded mostly from tamarind and less frequently from baskets and gunny sacs used to carry fishes.

***Carpophilus hemipterus* (Linnaeus)**

Dermestes hemipterus Linnaeus, 1758. *Syst. Nat.*, ed. 10: 358.

Carpophilus hemipterus: Stephens, 1830, *Illus. Brit. Ins.*, 3: 50.

Diagnosis

Dull or shiny black, with two conspicuous, amber-brown spots at posterior tips of elytra and two smaller, more obscure spots of same colour near bases of lateral margins of elytra; antennae and legs amber coloured; surface of body is finely punctate, each pit gives rise to a hair (Fig. 1(3)).

Size: 3 mm.

Material studied

30 exs. India: Kolkata, Ultadanga, ex. keora and mahua seeds, 14.v.1983; P. K. Basak; 20 exs, Kolkata, Shyambazar, ex. groundnut seeds, 19.iii.1983; P. K. Basak; 10 exs; Kolkata, Mechubazar, ex. walnuts, 9. iv.1983; P. K. Basak; 7 exs; Kolkata, Sovabazar, ex. maize grain, 19.iii.1982; P. K. Basak; 3 exs.

Distribution

World-wide; occurring throughout tropical and temperate parts of the world (Hinton, 1945). From India this species has been recorded from West Bengal and Bihar.

Remarks

Hinton (1945) recorded this species from grapes, raw sugar, figs, cloves, copra, dried apples, apricots, prunes, avocado, raisins, bananas, limes, peaches, plums, dates, oranges, pears, melons, tomatoes, honey, sorghum, pineapple, corn, wheat, rice bran, corn meal, cotton seeds, oats, shelled peanuts, and other nuts. This species also feeds on the pulp of the fruit itself and infests readily if contaminated with fungus. Normally they cannot enter the fruit unless the surface is broken or damaged. It was also found, in less number, on cereals, oilseeds and their products. They infest mostly 'keora' seeds, mahua and groundnut seeds and less so walnuts and maize grain.

Carpophilus marginellus Motschulsky

Carpophilus marginellus Motschulsky, 1858, *Etud. Ent.*, 7: 40.

Diagnosis

Dark brown, dorsally and underside reddish brown, strongly punctate, covered by recumbent golden hairs (Fig. 1(4)).

Size: 2.2–3.5 mm.

Material examined

20 ex. India: Howrah, Railway yard, *ex. bentonite powder*, 24.iv.1983; P. K. Basak; 5 exes., Kolkata, Sealdash Railway yard, *ex. portland cement*, 4.viii.1983; P. K. Basak; 2 exs., Kolkata, Sealdah, *ex. tamarind*, 15.vii.1983; P. K. Basak; 4 exs., Kolkata, Burrabazar, *ex. wheat bran*, 26.vi.1982; P. K. Basak; 3 exs., Kolkata, Posta, *ex. flour*, 24.iv.1985; P. K. Basak 1 ex., Kolkata, Sovabazar, *ex. maize grain*, 19.iii.1982; P. K. Basak; 5 exs.

Distribution

Widely distributed in tropical and sub-tropical parts of the world, including USA, Britain, W. Africa, Madagascar, Sri Lanka, Indo-China, Malaysia, Hong Kong, Japan, Pacific Islands, and presently recorded from India (W. Bengal) for the first time.

Remarks

Dobson (1954a,b) recorded this species from cocoa beans in W. Africa, and wheat from Maryland, USA. Aitken (1975) recorded it from wheat, maize, cocoa beans, soya beans, desiccated cocoanut, copra and what from different countries, and Hinton (1945) encountered it in rice and flour. In the present study, it was collected mostly from bentonite powder, moderately from tamarind, wheat bran, flour, maize grain, and occasionally from portland cement. The specimens collected from mango seeds (ZSI) have also been examined.

Carpophilus obsoletus Erichson

Carpophilus obsoletus Erichson, 1843. *German Zeitschr. Fur. Ent.* 259.

Diagnosis

Blackish; elytra dark brown with pale humeral angles; sparsely pubescent (Fig. 1(5)).

Size: 2.3–3.8 mm.

Material examined

9 exs. India: Kolkata, posta, *ex.* flour, 24.iv.1985; P. K. Basak; 2 exs., Kolkata, Sovabazar, *ex.* maize grain, 19.iii.1982; P. K. Basak; 3 exs., Kolkata, Ultadanga, *ex.* mahua seeds, 14.v.1983; P. K. Basak; 2 exs., Kolkata, Shyambazar, *ex.* groundnut seeds, 19.iii.1983; P. K. Basak; 1 ex.

Distribution

Widespread throughout the tropics and sub-tropics; found in Europe, Asia, Africa including Madagascar, India, Sri Lanka, Phillipines, Malaysia, Japan, Formosa, Iraq, Solomon islands, S. America, West Indies. In India, this species is known from West Bengal, Bihar, Arunachal Pradesh, Maharashtra and Andaman Islands.

Remarks

This is a widespread pet of grains, grain products, oilseeds, copra, oilcakes, cocoa beans, dried fruits, nuts, illipenuts, and sago flour (Aitken, 1975). Sengupta *et al.* (1984) recorded it from dried plums. In the present study, this species was observed mostly in flour and maize grains, and moderately in mahua, groundnut seeds, coir etc.

Carpophilus pilosellus Motschulsky

Carpophilus pilosellus Motschulsky, 1758, *Etud. Ent.* 7: 41.

Diagnosis

Reddish brown, surface shiny with recumbent pubescence (Fig. 1(6)).

Size: 1.8–2.9 mm.

Material examined

5 exs. India: Kolkata, Ultadanga, *ex.* keora seeds, 14.v.1983; P. K. Basak; 2 ex., Kolkata, Shyambazar, *ex.* groundnut seeds, 19.iii.1983; P. K. Basak; 1 ex., Kolkata, Mechubazar, *ex.* fruit basket, 9.iv.1983; P. K. Basak; 1 ex., Kolkata, Howrah, *ex.* fish baskets and gunny sacs, 15.i.1982; P. K. Basak; 1 ex.

Distribution

China, Japan, Madagascar, Indo-China, Sulawesi, Micronesia. From India this species has been reported from Delhi, Jharkhand and West Bengal.

Remarks

This species was recorded from ripe banana, copra, chicken feed, decaying cocoanut, leaf litter, fodder like plant, spider web and chocolate bar (Gillogy, 1962). Aitken (1975) collected it from rice, rice bran, wheat mixed with maize, brazil nuts, maize meal, oilcakes, gallnuts and illipenuts. In the present study, it was collected mostly from decaying keora seeds, and occasionally from groundnuts, fish scales, fruit baskets, and gunny sacs used for carrying fish.

KEY TO SPECIES OF *CARPOPHILUS* STEPHENS FROM STORED PRODUCTS

1. Apex of pygidium bluntly rounded 2
- Apex of pygidium points 3
2. Pronotum 1.5x wider than long, broadest in middle, sides feebly arcuate, without basal foveal; antennal segment 3 as long as segment 2 (Fig. 1(2)) *Carpophilus freemani* Dobson.
- Pronotum slightly wider than long, broadest in basal half, sides curved uniformly in anterior half, with basal foveae; antennal segment 3 shorter than segment 2 (Fig. 1(3)) *Carpophilus hemipterus* (L.)
3. Posterior margin of pronotum distinctly curved; antennal segment 3 shorter than or as long as segment 2 4.
- Posterior margin of pronotum feebly sinuate; antennal segment 3 slightly longer than segment 2 (Fig. 1(4)) *Carpophilus marginellus* Motschulsky.
4. Propygidium and pygidium elongate, elytra feebly transverse with no depression at base 5.
- Propygidium and pygidium short and transverse; elytra much wider than long and depressed at base (Fig. 1(1)) *Carpophilus delkeskampi* Hisamatsu.
5. Body oblong-oval, not shining, elytra blackish-brown with pale humeral angles; antennal club with rounded apex (Fig. 1(5)) .. *Carpophilus obsoletus* Erichson.
- Body oblong, shining, elytra uniformly reddish brown; antennal club with acuminate apex. (Fig. 1(6)) *Carpophilus pilosellus* Motschulsky.

DISCUSSION

A large number of beetles are of considerable economic importance as they infest and damage a wide range of stored food and other commodities of biotic origin. At least 500 species have hitherto been listed as primary infesters of many plant and animal products in stores and warehouses from the world. Many more species are encountered in storage and in packing material as associates which are otherwise scavengers, predators or mould feeders. All these species have specialized ecological requirements and life patterns that influence the degree of economic loss. Identification of these beetles is quite often not easy. A survey of storage products in Kolkata was therefore undertaken to study the species found in these habitats and at least 85 species have

been found. This study of *Carpophilus* species resulted in recording 5 more species from the storage sites in India.

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Impact of IPM on natural enemies in irrigated cotton of North India

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ABSTRACT: Large-scale farmers' participatory field validation trials were conducted at village Panihari in Sirsa district of Haryana for 3 crop seasons during 2002–04 to evaluate impact of Integrated Pest Management (IPM) on conservation of natural enemies in irrigated cotton. IPM module consisted mainly of balanced fertilizer use, regular scouting and pheromone monitoring, need based application of neem products alternating with chemical insecticides and release of *Beauveria bassiana* and *Trichogramma chilonis*. The Farmers' Practices (FP) relied exclusively on chemical insecticides and use of higher/lower fertilizer dose than the recommended dose. The average number of chemical insecticidal sprays was substantially reduced to 2–3 sprays in IPM against 6–7 single or mixed sprays in FP. The overall reduction of pesticide use over three seasons reflected in high population of natural enemies i.e. 0.35 eggs + larvae of *Chrysoperla carnea* and 0.16 adults of spiders per plant in IPM compared to 0.14 eggs + larvae of *C. carnea* and 0.07 adults of spiders per plant in FP. Better parasitization of *Helicoverpa armigera* eggs by *T. chilonis* was in IPM block (22.4%) as compared to natural parasitization (2.2%) in FP.

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KEYWORDS: Integrated pest management, *Helicoverpa armigera*, *Trichogramma chilonis*, conservation, farmers' practices

INTRODUCTION

Indiscriminate use of broad spectrum insecticides to contain the insect pests in cotton ecosystem has very often resulted development of high level of resistance in bollworms, resurgence of sap sucking insects (Dhawan and Simwat, 1997) and destruction of beneficial natural enemies. Though significant progress has been made in central and south India to reduce pesticide load by popularizing IPM technology in rainfed cotton among farmers (Singh *et al.*, 2002), management of insect pests

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in North India is still largely based on the use of chemical insecticides, which has resulted in the wiping out of natural enemies. Attempts have also been made under Punjab conditions to develop and evaluate bio intensive IPM module in small plots (Verma and Gill, 1992; Brar *et al.*, 2002). During 2002–2004 large-scale farmers' participatory field trials were conducted on synthesis and validation of IPM in irrigated cotton at village Panihari (District Sirsa, Haryana) by National Centre for Integrated Pest Management (NCIPM), New Delhi.

MATERIALS AND METHODS

Field trials were conducted at farmers' fields in two blocks of 2 ha each in 2002 and 2003 and 8 ha each in 2004 to study the impact of IPM on population of important insect pests and natural enemies in irrigated cotton at village Panihari (Sirsa, Haryana). One block involved the common farmers' practices (FP) prevailing in the area to contain the insect pests whereas the other block involved the IPM practices. During 2002 IPM as well as FP included *Desi* cotton (*Gossypium arboreum*) variety, RG-8 and American (*G. hirsutum*) variety, H-1117 with standard agronomic practices. During 2003 and 2004 the trial was conducted with cultivar H-1098 in both IPM and FP blocks. In FP the farmers applied higher or lower dose of fertilizer than the recommended dose i.e. 70–80 kg urea/acre. The FP consisted entirely of sprays of chemical insecticides either alone or in mixtures whereas the IPM involved alternating application of 5% Neem seed powder extract (NSPE) or neem oil (2.5 l/ha) with 2–3 sprays of chemical insecticides (Table 1). The application of chemical insecticides was based upon the insect pests prevailing in the season; therefore, the schedule of chemical insecticides did not remain same during three years. In addition IPM also included the application of *Beauveria bassiana* @ 1.25 kg/ha (4×10^8 CFUs), 1 or 2 releases of egg parasitoid *Trichogramma chilonis* @ 150 000 ad/ha in August/September and installing 5 pheromone traps/ha from July to September in the field to monitor the population of *Helicoverpa armigera*. Field releases of *T. chilonis* was carried out when 2–3 moths *H. armigera* were trapped in each pheromone trap per night for consecutive 3–4 days or the eggs of *H. armigera* were available in the fields. A gap of one week before and after the release of *T. chilonis* and application of chemical insecticide was maintained. The details of different interventions have been indicated in Table 1.

Observations on population of egg and larvae of *Chrysoperla carnea* and adults of spiders were recorded at weekly interval per plant by randomly selecting 25 plants/acre. The data were analyzed statistically using paired 't' test. To record observation on parasitisation of *H. armigera* eggs by *Trichogramma*, the eggs of *H. armigera* were collected from FP and IPM blocks in the month of September and were transferred individually in glass vials (7.5 × 2.5 cm) under optimum laboratory conditions ($25 \pm 2.0^\circ\text{C}$ and 60% RH) for emergence of parasitoid or host larvae.

TABLE I. Components of IPM and Farmers' practices (FP)

Month	2002			2003			2004		
	IPM	FP	IPM	FP	IPM	FP	IPM	FP	IPM
July	NSPE + 200 ml/ha NSPE 2. Imidacloprid 125 ml/ha NSPE	1. Triazophos 1.5 l/ha + Alphamethrin 200 ml/ha			1. Endosulfan 2.0 l/ha + Dichlorvos 750 ml/ha		Neem oil 2.5 l/ha <i>B.bassiana</i>		1. Meothrin 200 ml/ha 2. Triazophos 400 ml/ha + Alphamethrin 200 ml/ha
Aug.	1. Alphamethrin 200 ml/ha NSPE 2. Imidacloprid 125 ml/ha NSPE	2. Quinalphos 2.5 l/ha Fenvalerate 500 ml/ha	NSPE <i>B.bassiana</i>	1. Quinalphos 2.5 l/ha Fenvalerate 500 ml/ha	2. Imidacloprid 125 ml/ha 1 l/ha	1. Profenophos 2.5 l/ha	3. Profenophos 2.5 l/ha+ Meothrin 200 ml/ha		
	3. Quinalphos 2.5 l/ha	4. Monocrotophos 750 ml/ha + Fenvalerate 500ml/ha 5. Indoxacarb 500 ml/ha	NSPE		3. Lambdacyhalothrin 400 ml/ha + Neomexcel 4. Acephate 400 ml/ha + Alphamethrin200 ml/ha	3. Lambdacyhalothrin 400 ml/ha + <i>T. chilonis</i> (I release) 2. Indoxacarb 500 ml/ha	Neem oil 2.5 l/ha; <i>T. chilonis</i> (I release) Indoxacarb 500 ml/ha		
Sept.	NSPE + <i>B. bassiana</i>	6. Ethion 1.0 l/ha +Fenvalerate 500 ml/ha		2. Acephate 400ml/ha 3. Alphamethrin 200 ml/ha; <i>T. chilonis</i> Two sprays pf NSPE at 10 days interval	5. Triazophos 400 ml/ha + Meothrin 200 ml/ha 6. Indoxacarb 500 ml/ha 7. Acephate 400 ml/ha + Fenpropathrin 200 ml/ha	5. Triazophos 400 ml/ha + Meothrin 200 ml/ha 6. Indoxacarb 500 ml/ha 7. Acephate 400 ml/ha + Fenpropathrin 200 ml/ha	4. Indoxacarb 500 ml/ha+Acetamiprid 125 gm/ha 5. Indox- acarb 500 ml/ha + Meothrin 200 ml/ha 6. Indoxacarb 500 ml/ha		

NSPE-Neem Seed Powder Extract (12.5-25.0 Kg/ha as per stage of the crop);

Bb.- *Beauveria bassiana* @ 1.25kg/ha (4×10^8 CFUs); *T. chilonis* @ 150 000 ad/ha

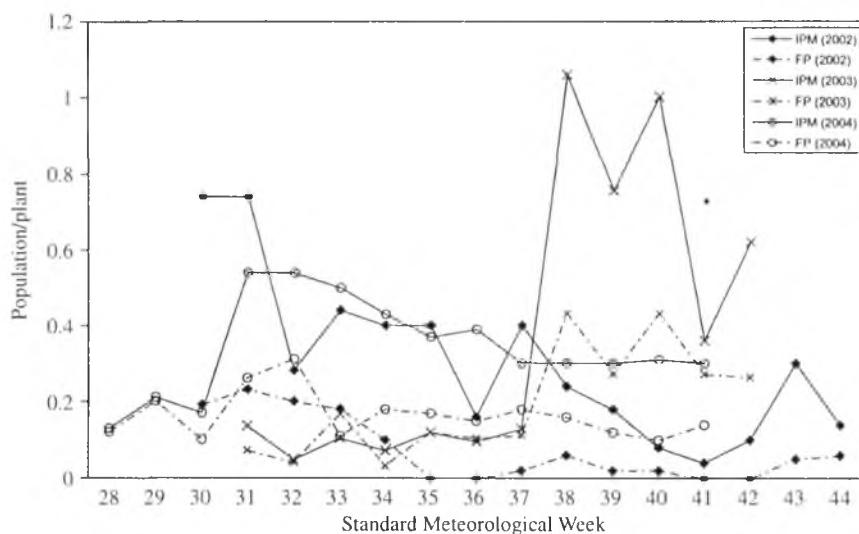


FIGURE 1. Population of *Chrysoperla carnea* (eggs) in IPM and Farmers' Practices (2002–04).

RESULTS AND DISCUSSION

As many as 274 species of biological control agents mainly arthropod parasitoids (162) and predators (90) have been reported (Lingappa *et al.*, 2001) but only a few bio-agents are available throughout the season in North India. In the present investigation the study was focused on the population dynamics of *C. carnea*, *Trichogramma* spp. and spiders during three consecutive years. As per the results for three years (Table 1), the average eggs and larval population of *C. carnea* remained 0.35/plant in IPM against 0.14/plant in FP. The differences were statistically significant in 2003 and 04. Weekly observations indicated highest population of the predator on 30th and 31st (0.7 eggs/plant), 38th (1.06 eggs/plant) and 31st and 32nd (0.54 egg/plant) Standard Meteorological Weeks (SMW), during 2002, 2003 and 2004, respectively (Fig. 1). During 2002 and 2004, after the initial high population build-up in 31st/32nd SMW, there was a decline in the population of eggs and larvae of *C. carnea* both in IPM and FP blocks. In FP 6–7 sprays of insecticide mixture were done (including 10–12 chemical insecticides), compared to 2–3 sprays in IPM, therefore, the reduction in the population in FP was very high and reached to a level of zero in 35th and 36th SMW in 2002 (Fig. 1). Among the three years the highest population (>1 egg or larvae/plant) was observed in 38th Standard Meteorological week (SMW) in September, 2003. Studies made at Guntur under the field conditions on the effect of common as well as new insecticides of different origin against whitefly in cotton fields indicated significant reduction in the populations of coccinellids and *Chrysopa* compared to control (Venugopal Rao *et al.*, 1990). High level of toxicity of carbaryl (Hassan *et al.*, 1987), monocrotophos and quinalphos (Singh and Verma, 1986; Srinivasan and

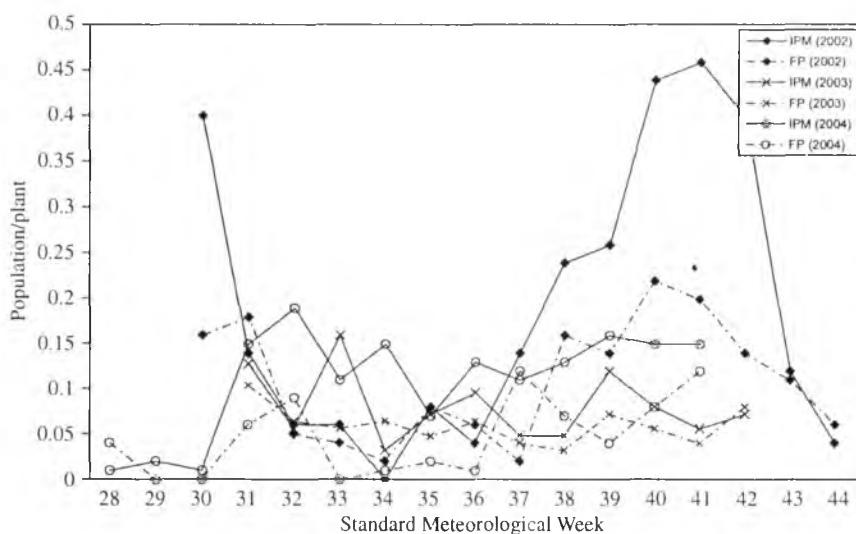


FIGURE 2. Population of spiders (adults) in IPM and Farmers' Practices (2002–04).

TABLE 2. Population (number/plant) of *Chrysoperla carnea* and spiders in IPM and Farmers' fields (FP)

Block	<i>Chrysoperla carnea</i> (eggs+larvae)				Spiders (Adults)			
	2002	2003	2004	Mean	2002	2003	2004	Mean
IPM	0.31	0.38	0.35	0.35	0.19	0.08	0.11	0.13
FP	0.08	0.19*	0.16*	0.14	0.11	0.06	0.05*	0.073

*Paired 't' test significant at $p=5\%$

Sundara Babu, 2000) and most of organophosphate insecticides (Toda and Kashio, 1997) to *C. carnea* have also been reported. Nymphs of *C. carnea* have a remarkable tolerance of pyrethroids, attributable to detoxification by pyrethroid esterase (s) (Ishaaya and Casida, 1981).

In case of adult spiders variation in population was recorded in each year and the population, in general, remained higher in IPM as compared to FP (Table 2). Low population of spiders in FP could be due to more application of chemical insecticides which are having negative effect on the population densities and species diversity of field spiders (Paik and Hwang, 1990; Lee *et al.*, 1993). Results of three years indicated highest population of spiders in IPM during 2002. In this year there were two peaks i.e. 30th and 41st SMW which showed higher population of adult spiders i.e. 0.40 and 0.46 adult/plant, respectively (Fig. 2). Among different spiders the prominent spiders observed were lynx spider, *Oxyopes javanus* (Thorell); orb spider, *Argiope minuta* (Karsh); wolf spider, *Lycosa pseudoannulata* (Boesenbergh and Strand); long-jawed

TABLE 3. Parasitization of *H. armigera* eggs in IPM and Farmers' field (FP)

Block	Total <i>H. armigera</i> eggs collected			Eggs showed emergence of <i>H. armigera</i> larva (%)			Eggs remained unhatched (%)			Eggs parasitized by <i>Trichogramma</i> sp. (%)			
	2003		2004	Total	2003	2004	Mean	2003	2004	Mean	2003	2004	Mean
	IPM	76	97	173	47.4	43.5	45.4	31.5	32.8	32.1	21.1	23.7	22.4
FP	83	107	190	38.5	35.7	37.1	58.9	62.5	60.7	2.6	1.8	2.2	

spider, *Tetragnatha javana* (Thorell); *Neoscona theisi* (Walcknear) and *Peucetia viridana* (Stoliczka).

The results on parasitisation of *H. armigera* eggs by *Trichogramma* sp. indicated 21.2 and 23.7 per cent parasitisation in IPM block against 2.1 and 1.8 per cent in FP in 2003 and 04, respectively (Table 3). The increase in the egg parasitisation by *Trichogramma* sp. could be due to field release of *T. chilonis* (one release in 2003 and two releases in 2004) and less number of chemical insecticides in IPM as compared to FP. The neem products applied in the present study have also been reported to be safe to parasitoids and predators and non-insect predators (Singh and Singh, 1996). The percentage of unhatched eggs of *H. armigera* (Table 3) also remained high in FP than IPM. This could be due to ovicidal action of many pesticides used in FP. Under the Punjab conditions Dhawan and Simwat (1997) observed that the parasitisation of *Helicoverpa* eggs declined from 4.6 in 1975 to only 0.78 per cent in 1995. In the same state recent studies conducted on evaluation of bio-intensive module (Brar *et al.*, 2002) indicated 10.7 % parasitisation of *Helicoverpa* eggs in bio-intensive module against 0.7% in insecticidal treatment.

It is evident from the above results that field application of chemical insecticides can be reduced to a greater extent by use of *neem* based plant products and field releases of bioagents. IPM tactics have definitely helped in conservation of natural enemies in irrigated cotton ecosystem of North India where the natural enemies have been severely affected by repeated large scale of exclusive use of chemical insecticides.

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Super parasitism in Indian lac insect, *Kerria lacca* (Kerr) and its implication on fecundity and resin producing efficiency of its two strains

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ABSTRACT: Parasitoids of lac insect inflict severe damage to the crop affecting adversely the resin yield and the fecundity of the insects, particularly during rainy season. There was 18.40% in *kusmi* and 26.00% parasitisation in *rangeeni* strain (female insects); *Aprostocetus purpureus* alone caused 7.8 and 11.8% parasitisation respectively. Up to nine parasitoids in *rangeeni* and six in *kusmi* were found from a single cell. Average reduction in resin produced by a single female due to parasitism varied between 17.25–39.80% in *rangeeni* and 25.24–37.91% in *kusmi* strain whereas, reduction in fecundity of lac insects ranged between 22.44–96.82% and 25.29–90.39% respectively for the two strains. As the number of parasitoids in each cell increased, there was a corresponding decrease in resin production and fecundity; the latter being affected more severely. © 2007 Association for Advancement of Entomology

KEYWORDS: Indian lac insect, *Kerria lacca*, resin-yield, fecundity, super parasitism, *Aprostocetus purpureus*

INTRODUCTION

Kusmi and *rangeeni* are two strains of Indian lac insect *Kerria lacca* (Kerr) which are commonly exploited for lac cultivation. Of the many problems confronting lac culture, the most serious, undoubtedly is the problem of management of pests and diseases of lac insect. Insect enemies of Indian lac insect, *Kerria lacca* (Kerr) cause considerable damage to the lac crop. The earliest reference on parasitoids of lac insects is of Stebbing (1910) who figured three, while Varshney (1976) has reported thirty different insects as parasitoids of lac insects. Two more parasitoids have been reported after that (Rizvi, 1986). Though lac predators are known to inflict severe damage to lac crop, losses due to parasitisation are also not insignificant. However, extent and

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nature of damage caused by parasites is not well documented. Whatever little has been done, pertains mostly to extent of parasitisation and relative/seasonal abundance of parasitoids associated with lac insect (Glover, 1937; Srivastava *et al.*, 1984; Srivastava and Mehra, 1984). Also, Majumdar *et al.* (1962) showed interrelationship amongst population of parasitoids of lac insect, and Chowdhury *et al.* (1971) worked out relationship between density of lac insect and its parasitoids.

Fecundity of lac insect and the resin secreted by it are two important economic parameters; the former being important for propagating next crop while the later is product of commerce. Sharma and Ramani (2001) observed parasitoids reduce yield and fecundity. Instances of super parasitism are not uncommon in lac insects. *Aprostocetus purpureus* Cam. (Eulophidae) is one of the most abundant parasitoids associated with lac insects. Hence, in the present study effect of super parasitism caused by *A. purpureus* on fecundity and resin producing efficiency of Indian lac insect, *Kerria lacca* (Kerr) has been evaluated.

MATERIALS AND METHODS

Mature individual female lac insect cells at crop maturity were collected from the Institute Research Farm from lac crops on *Butea monosperma* (*palas*) for *rangeeni* and *Schleichera oleosa* (*kusum*) for *kusmi* strain during rainy season generation as lac culture is more vulnerable to parasitoids in this season. 500 cells from each host-plant were collected. Recommended package of practices for lac cultivation was followed. The cells were put individually in five ml glass vials and then plugged with cotton. After a month of storage, emergence of lac larvae and parasitoids was completed. Then the cells were grouped into healthy and parasitized ones based on the presence of small neatly cut hole(s) in the cells by emerging parasitoid(s) and/or presence of the parasitoid(s) in the cell/vial. Cells parasitized only by *Aprostocetus purpureus* were considered for grouping in to separate classes based on the number of parasitoids emerged for the present study.

Size, weight of lac secreted and fecundity of cells belonging to each class were measured after a month of storage. Since female cells are globular in shape, average of horizontal and vertical diameter measured with the help of a screw gauge was taken as cell size. Total number of larvae that emerged from the female lac insects in the vial and the larvae which could not emerge were counted after breaking the cell open. The whole count was used as fecundity of the female. Dead female insect was removed and weight of resin secreted by individual cell was recorded using an electronic balance. Healthy cells were also grouped into corresponding different classes on the basis of their size. Cells not conforming to any of the class in parasitized population were not considered for the study. Mean value of each class was used to compare various parameters of healthy and parasitized cells by paired '*t*' test (Panse and Sukhatme, 1985).

RESULTS

Extent of parasitisation

Rangeeni strain of lac insect was more vulnerable to parasitoids (26.0%) than *kusmi* (18.4%). Among all the recorded parasitoids of lac insect, *Aprostocetus purpureus* alone caused more than 40% parasitization in both the strains (7.8 and 10.8% respectively in *kusmi* and *rangeeni* strains). Sharma *et al.* (1997) have also recorded similar frequency of relative parasitism in lac insect. Instances of super parasitism were recorded from both the strains. Up to nine parasitoids were recorded from a single cell in *rangeeni* and six in *kusmi* strain. Frequency of occurrence of parasitized cells was indirectly related to the number of parasitoids in a cell.

Effect of super parasitism on resin yield and fecundity in *rangeeni* strain

Though mean diameter of the parasitized cells did not differ significantly from that of healthy cells, resin secreted by a parasitized lac insect was 23.38% less than the healthy cells. As the number of parasitoids in a cell increased, there was a proportional and significant decrease in the resin secreted as well as fecundity of the insect. The effect is more pronounced on fecundity. Mean fecundity per cell in a healthy insect was 325.74 in comparison to 197.78 in parasitized cells. Decrease in fecundity was recorded between 22.44%–96.82% depending upon the number of parasitoids in the cells (Table 1).

Effect of super parasitism on resin yield and fecundity in *kusmi* strain

Weighted mean of all the parameters scored viz., diameter of the cell, weight of the cell and fecundity were higher than the *rangeeni* strain. Difference in the mean diameter of the parasitized and healthy cells was insignificant, but mean of the resin secreted by an insect affected by the parasitoid was 14.567 mg which was 23.24% less than the resin secreted by the healthy insects (Table 2). Similarly, fecundity was affected much more adversely. Mean fecundity of healthy insects ranged between 281–427 in different size classes and that of parasitized cells 27–319. With increase in number of parasitoids in a cell, resin secreted by the insect and its fecundity also decreased.

DISCUSSION

Thickness of lac encrustation is one of the criteria for assessing the quality of broodlac. It is evident from the data that size of parasitized as well as healthy cells did not differ much but comparison of weight of the resin secreted by the two revealed that the amount of the resin produced by the parasitized cells was significantly lower. Hence, visual assessment of broodlac quality on the basis of encrustation thickness alone may prove to be deceptive unless weight is also taken into account.

Female lac insects are not only chief lac producers but they also serve to propagate next crop as broodlac (analogous to seed in other crops). Any adverse effect on fecundity of lac insect would have far reaching implications on the next crop. Mehra

TABLE I. Effect of super-parasitism on fecundity and resin yield of the *rangeeni* strain of Indian lac insect, *Kerria lacca* (Kerr) during rainy season crop

Parasitoids recorded from a single cell	Cells parasitized by <i>Aprostocetus purpureus</i>						Healthy cells					
	Frequency	Mean Diameter (mm)	Mean Weight (mg)	% reduction of resin over healthy cells	Mean Fecundity	% reduction over healthy cells	Frequency	Class	Mean Diameter	Mean Weight	Mean Fecundity	
1	21	3.42	10.60	17.25	280	22.44	39	3.41-3.50	3.43	12.81	361	
2	15	3.34	9.43	24.80	206	43.09	42	3.31-3.40	3.36	12.54	362	
3	10	3.27	8.16	33.37	155	55.71	57	3.21-3.30	3.24	12.37	350	
4	6	3.07	7.69	35.92	112	65.85	50	3.01-3.10	3.06	11.74	311	
5	3	3.15	7.50	35.90	97	68.81	72	3.11-3.20	3.16	12.00	328	
6	1	2.92	6.89	37.71	56	79.78	35	2.91-3.00	2.95	11.25	299	
7	1	2.77	7.06	37.24	93	72.24	11	2.71-2.80	2.74	10.82	215	
8	1	2.69	6.01	39.80	7	96.82	5	2.61-2.70	2.66	9.98	220	
9	1	2.86	6.70	37.99	30	86.05	21	2.81-2.90	2.85	10.99	277	
Weighted Total	59	193.73	540.95	-	11669	-	332	-	1042.19	3972.97	108146	
Weighted Mean	-	3.284 ^{ns}	9.169**	23.38	197.78**	39.28	-	-	3.153	11.967	325.74	

ns - non significant, ** = $P < 0.01$

TABLE 2. Effect of super-parasitism on fecundity and resin yield of the *kusmi* strain of Indian lac insect, *Kerria lacca* (Kerr) during rainy season crop

Parasitoids recorded from a single cell	Cells parasitized by <i>Aprostocetus purpureus</i>					Healthy cells					
	Frequency	Mean Diameter (mm)	Mean Weight (mg)	% reduction of resin over healthy cells	Mean Fecundity	% reduction over healthy cells	Frequency	Class	Mean Diameter of the cells	Mean Weight of the cells (mg)	Mean Fecundity
1	17	3.96	15.7	25.24	319	25.79	55	3.91-4.00	3.93	21.0	427
2	9	3.85	15.0	27.54	300	27.18	61	3.81-3.90	3.84	20.7	412
3	8	3.70	13.8	29.59	212	45.50	94	3.71-3.80	3.74	19.6	389
4	3	3.64	11.9	33.15	132	60.83	50	3.61-3.70	3.65	17.8	337
5	1	3.47	9.5	37.91	27	90.39	32	3.41-3.50	3.46	15.3	281
6	1	3.52	10.6	36.90	81	73.18	45	3.51-3.60	3.55	16.8	302
Weighted Total	59	149.48	568.1	-	10323	-	337	-	1254.92	6395.7	124615
Weighted Mean	-	3.833 ^{ns}	14.567 ^{**}	23.24	264.69 ^{**}	28.42	-	-	3.724	18.978	369.78

ns - non significant, ** = $P < 0.01$

and Majumdar (1963) have studied emergence of larvae from parasitized female lac cells and concluded that emergence of larvae is not always hampered by parasitisation which is contrary to the observations of the present study, where it was found that decrease in fecundity of lac insect due to parasitisation ranged between 25.79–90.39 in *kusmi* and 22.44–96.82% in *rangeeni* strain. Thus, proportionately more broodlac would be required as compared to healthy broodlac for inoculating the same number of trees. If parasitized at an early stage, the lac insect is practically eaten up by the developing parasitoid rendering the lac useless for broodlac purpose. Moreover, the broodlac harbouring parasitoids if used for raising next crop would serve as source of infection to new lac culture.

Lac insects which are parasitized at an early stage die before reaching maturity. Also, Jaiswal and Saha (1995) have found positive and significant correlation between density of lac insects and number of parasitoids. Since the present study was confined only to isolated and adult female lac insects, it is highly likely that extent of parasitisation in continuous encrustation of lac insects under field conditions would be higher than recorded causing more damage.

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New species of *Thelcticopis* (Araneae: Sparassidae) from India

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ABSTRACT: A new species of spider belonging to the genus *Thelcticopis*, namely *Thelcticopis moolampilliensis* sp. nov., is described and illustrated from Kerala, India.

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KEYWORDS: *Thelcticopis*, spider, Kerala, Sparassidae

INTRODUCTION

Thelcticopis is a small oriental genus of large huntsman spiders. This genus is characterized by the presence of 5–6 small teeth on the inner margin of chelicerae and 5–7 pairs of ventral spines on tibia of first leg. Spinnerets in this genus are supported on a membranous stalk and strengthened by a hairy chitinous ring. Pioneering studies on Indian *Thelcticopis* was done by Pocock (1901). He described four species from India namely *T. ajax* Pocock, 1901 from Ootacamund, *T. bicornuta* Pocock, 1901 from Naga Hills, *T. rufula* Pocock, 1901 from Nilgiri Hills, and *T. virescens* Pocock, 1901 from Trivandrum. Later *T. maindroni* Simon, 1906, *T. canescens* Simon, 1887, *T. serambiformis* Strand, 1907, *T. ancorum* Dyal, 1935 and *T. telonotata* Dyal, 1935, were reported from Indian region. Recently Sethi and Tikader (1990) published redescription of *T. canescens* Simon, 1887 and *T. maindroni* Simon, 1906. A new species of *Thelcticopis* is described and illustrated here. This is the only species of *Thelcticopis* recorded from Kerala other than *T. virescens* Pocock, 1901.

MATERIALS AND METHODS

Spiders were collected by methods described by Tikader (1987). Collected spiders were preserved in 70% ethyl alcohol and studied under Stereomicroscope. All measurements were taken with an eyepiece graticule. Epigyne was studied by clearing in 10% KOH. The status of the species was confirmed by referring to Pocock (1900,

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1901) and Barrion and Litsinger (1995); it is also confirmed at Zoological Survey of India, Kolkata. The type specimens are now at the Arachnological Collections of Department of Zoology, Sacred Heart College, Thevara; which will in due course be deposited at the National Collections of Zoological Survey of India, Kolkata.

Abbreviations used are as follows: AME = Anterior median eyes, ALE = Anterior lateral eyes, PME = Posterior median eyes, PLE = Posterior lateral eyes, L = Length, W = Width, H = Height.

RESULTS

Thelecticopis moolampilliensis sp. nov.

Colour: Dorsum of cephalothorax uniformly reddish brown without any patches, legs reddish brown, a little paler than cephalothorax. Labium, sternum, maxillae reddish brown. A slightly pale dorsal line on the cephalothorax. Chelicerae reddish, fang and teeth deep reddish brown. Eyes reddish yellow, bordered by black margin. Abdomen whitish on the dorsum, lateral edges bear dark brown, broad, marginal patches, inner end of these patches uneven. Mid dorsal line bears five dark brown spots; middle three spots are more broader than anterior and posterior spots. Spinnerets reddish brown. Ventrum of abdomen white with a moderately sized patch on the middle at about the centre of epigyne and spinnerets. Tarsal claws and teeth deep reddish brown. Scopulae on labium and maxillae reddish yellow. Leg spines deep reddish brown, pedipalp reddish brown.

In living specimens, the cephalothorax is reddish brown and the abdomen white in color.

Female

Cephalothorax: Longer than wide, clothed with white pubescence. Anterior 1/5th abruptly narrowed; front margin convex with thin hairs. Fovea fine, long, longitudinal. **Eyes:** Eight in two rows encircled by black margins; anterior eye row slightly shorter and procurved, AME slightly larger, and closer to each other. Posterior eye row longer and procurved, eyes widely separated, PME much closer to each other than to laterals. Ocular quadrangle longer than wide, narrower in front. **Sternum:** Longer than wide, heart shaped, reddish brown, paler towards anterior margin, anterior end truncated, posterior end pointed, clothed with hairs, broadest at coxae I & II, and narrower towards coxae III & IV. **Labium:** Similar in colour to sternum, longer than wide, anterior half narrowed and bluntly rounded, scopulae present. **Maxillae:** Longer than broad, broader apically, scopulae brown. **Chelicerae:** Strong, outer surface more convex; inner margin with five teeth and outer margin with three slightly larger teeth. **Legs:** Reddish brown, long and stout, clothed with hairs and few spines. Leg formula 1423. Femur I with two dorsal spines, femur II, III, IV with three dorsal spines. Tarsal claws two, with nine teeth in each claw. Femur of palp with four dorsal spines, tibia with five spines; femur longer than tibia. Tibia I with five pairs of ventral spines.

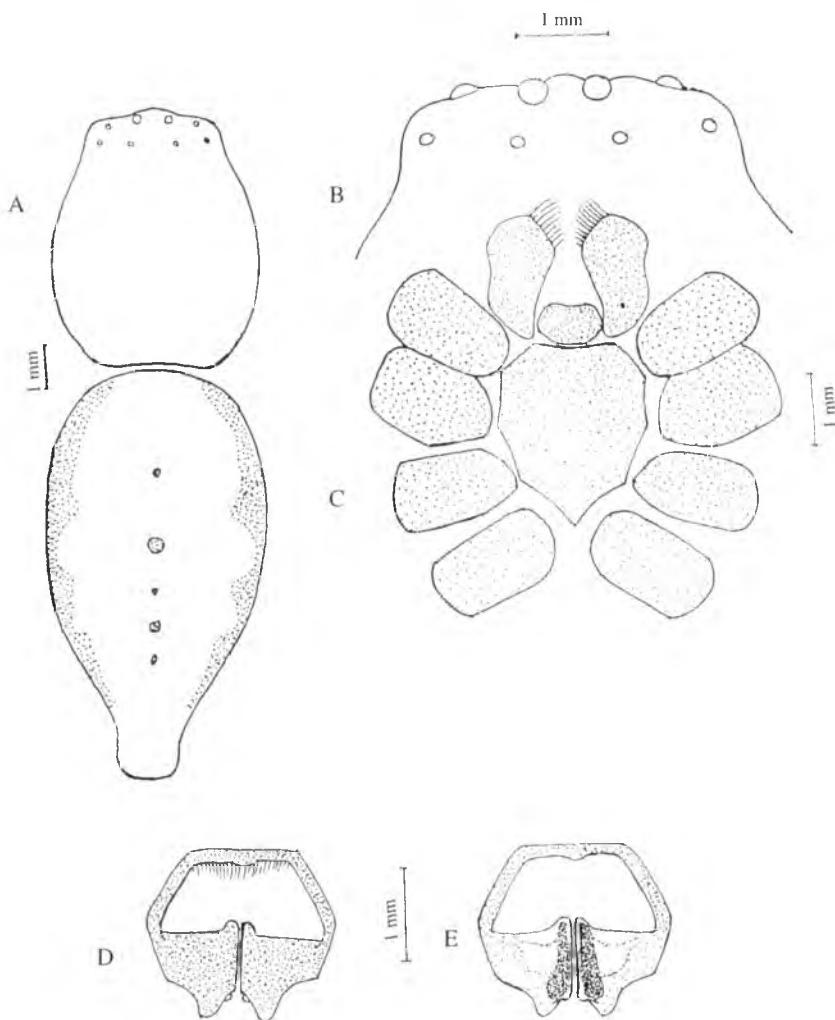


FIGURE 1. *Thelcticopis moolampilliensis* sp. nov. (female) A. Dorsal view of cephalothorax and abdomen, B. Eyes, C. Sternum with maxillae and Labium, D. Epigynum ventral view, E. Epigynum, dorsal view.

Metatarsus and tarsus I with ventral scopulae. *Palp*: Femur with three, tibia with four, and tarsus with three spines.

Abdomen: Longer than wide, narrowed posteriorly, widest at the middle. Two pairs of sigilla present. Ventrum pale, a large dark brown spot near to spinnerets.
Spinnerets: Supported by a hairy chitinous ring; anterior spinnerets widely separated.
Epigynum: With two lateral sclerotized pieces; posterior half of which are slightly

TABLE 1. Leg measurements for *Thelecticopis moolampiliensis* ♀ (mm)

Leg	Femur	Patella	Tibia	Metatarsus	Tarsus	Total
I	4.8	1.5	4.0	3.1	1.3	14.7
II	4.5	1.4	3.5	3.1	1.2	13.7
III	3.8	1.2	3.0	3.0	1.1	12.1
IV	4.0	1.2	4.0	3.5	1.2	13.9

separated in the middle, anterior margin bordered by a narrow sclerotized margin. A widely depressed, open cavity present in the anterior half, partly divided by median segment in the middle.

Measurements: (Table 1) Total length: 17 L, Cephalothorax 6 L, Abdomen 10 L.

Ocular measurements: Eye diameter: AME = 0.36, ALE = 0.252, PME = 0.144, PLE = 0.18. Eye separation: AME-AME = 0.288, AME-ALE = 0.432, PME-PME = 0.648, PME-PLE = 0.828.

Male (Immature)

Cephalothorax: Body pattern and colour similar to female. Clypeus narrow, less than half the diameter of AME. *Eyes:* Both rows of eyes straight or very slightly procurved, posterior row longer than anterior row. PME smaller than other eyes. AME nearer to each other than ALE. *Legs:* Femur I & II without ventral spines. Tibia I & II with five pairs of ventral spines. Metatarsus I with two ventral spines on each side, femur II with five dorsal spines. *Palp:* Tarsal claws with four teeth, tarsus with two spines on each side. *Chelicerae:* Outer margin with three large teeth, inner margin with five smaller teeth.

Abdomen: More yellowish than female, variegated with median and lateral brownish patches. median patch consists of five spots, and lateral patches also have five spots with posterior patches being contiguous. Ventrum pale yellowish, without the black patch seen in female.

Specimen examined: Holotype 1♀, Moolampilly Is., Kochi, Kerala, India, 30.xii.2000. Coll. Sunil Jose, K.

Paratype: 2♀, 1♂ immature, Kalamasserry, Kochi, 20.ii.2001, Coll. Sunil Jose, K.

Distribution

Moolampilly Island and Kochi, Kerala, India.

Natural History:

Collected from leaves of *Mangifera indica* at night (8 PM) found moving along the leaves.

Diagnosis: Dorsal surface of abdomen white, with lateral edges bearing dark brown broad marginal patches. Ventrum with a small isolated black patch present little below the middle. Chelicerae with inner margin bearing five teeth and outer margin bearing three slightly larger teeth. Leg 1 with five pairs of ventral spines.

Etymology:

The species is named after the type locality, Moolampilly Island, near Kochi.

Remarks:

T. moolampilliensis is closely similar to *T. rufula* Pocock, 1901, but can be separated by following differences:

1. Tibia of first pair of legs with five pairs of ventral spines in *T. moolampilliensis* as against six pairs of spines in *T. rufula* Pocock, 1901.
2. In *T. rufula* Pocock, 1901, ventral side of abdomen with a median dorsal band consisting of a series of black spots, whereas in *T. moolampilliensis* only a small isolated black patch present little below the middle.
3. Legs, sternum and coxa ochre yellow in *T. rufula* Pocock, 1901, whereas these are reddish brown in *T. moolampilliensis*.
4. Epigyne in *T. moolampilliensis* with two lateral sclerotized pieces in the posterior half and a widely depressed, open cavity in the anterior half. Female is not so far described in *T. rufula* Pocock, 1901. Collection of the immature male of *T. moolampilliensis* helped separating it from *T. rufula* Pocock, 1901.

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Activity pattern of juvenile hormone during metamorphosis in *Dysdercus cingulatus* Fabr. (Heteroptera: Pyrrhocoridae)

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ABSTRACT: JH being a morphogenetic hormone during nymphal stages, its titer at particular life stages determines which type of molt the developing insect has to undergo. Application of Juvenile hormone analogues on to 5th instar nymphs suggests that metamorphosis is a sequence of events each one of which is dependent on JH absence. © 2007 Association for Advancement of Entomology

KEYWORDS: juvenile hormone, methoprene, metamorphosis, *Dysdercus cingulatus*

Juvenile hormones are a homologous series of sesquiterpenoids that are involved in embryogenesis, molting, metamorphosis and reproduction (Gilbert *et al.*, 2000). In hemimetabolous insects metamorphosis involves wing growth, heteromorphic growth of segments of the legs and /or body segments. Further, the insect develops musculature necessary for flight and acquires the ability to respond to hormonal and or environmental cues involved in reproduction (Kumaran, 1990).

The insect *Dysdercus cingulatus* adults used for this study were reared in our laboratory under controlled conditions (temp: 28 + 31 °C, L/D cycle 12:12 and r.h 90 ± 3) and fed on soaked cotton seeds. Eggs were laid in clusters between cotton seeds and were collected at various intervals, and maintained so that enough number of nymphs and adults of known age were available for the present study.

Haemolymph was collected by cutting the antennae and draining hemolymph into an eppendorf tube. Tyrosinase activity was inhibited by the addition of a few crystals of phenylthiourea. It was then centrifuged at 10,000 rpm for 10 min at 4 °C in a refrigerated centrifuge (Universal 16 R, Hettich, Zentrifugen Germany) to remove the hemocytes and other debris. Supernatant was mixed with double the volume of sample buffer.

Abdomen was cut open; fat body was taken out and washed in Insect Ringer, homogenized in a pre-chilled homogenizer with homogenizing medium and centrifuged

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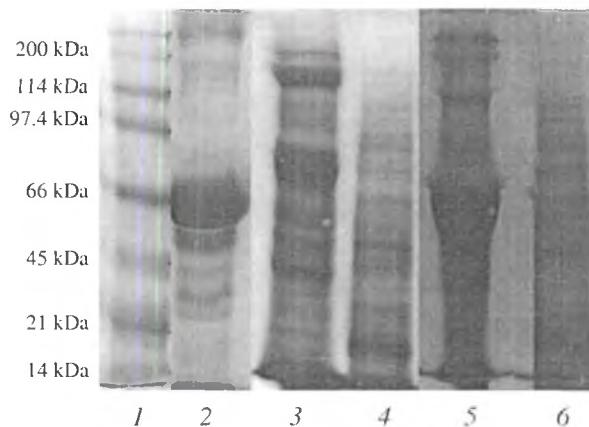


FIGURE 1. Electrophorogram showing changing protein profile in the hemolymph and fat body. Lane 1: Marker; Lane 2: 5th instar nymph hemolymph; Lane 3: 4-day-old adult hemolymph; Lane 4: Adult fat body; Lane 5: 0-day-old adult hemolymph; Lane 6: 5 th instar fat body.

at 5,000 rpm for 10 min at 4 °C. Supernatant was mixed with equal volume of sample buffer.

The electrophoretic protein profile of haemolymph was determined by one dimensional sodium-dodecyl-sulphate polyacrylamide gel electrophoresis (SDS-PAGE) carried out according to Laemmli (1970) under discontinuous and dissociating buffer systems. Gel electrophoresis was done using a vertical slab gel mini-electrophoresis unit (Genei, Bangalore) attached to power supply (LKB-2297 macro drives). One mm thick resolving gel (1.5 m Tris HCl pH 8.8) was prepared and stacking gel (0.5 m Tris HCl pH 6.8) was laid over the resolving gel.

Protein samples were mixed with sample buffer (50 mM Tris-HCl, pH 6.8, 2% (w/v) SDS, 10% (v/v) Glycerol, 0.1% Bromophenol blue, 5% 2-mercapto ethanol) such that the protein concentration of each sample was approximately 100 µg per well. The samples were boiled for 1–2 min in water bath maintained at 100 °C. The samples were then centrifuged at 5000 rpm for 10 min in a centrifuge to remove the debris 10 µl each was loaded on to the wells. Standard molecular markers were also run along with the samples. Electrophoresis was carried out initially at 60 v till the sample entered the resolving gel and at 120 v till the end of the run.

Juvenile hormone analogue, Methoprene (gift from Govindhan Bhaskaren, Texas A&M University, USA) was dissolved in acetone so as to get a concentration of 1 µg/µl solution. Methoprene was topically applied to the ventral region of 0-day, 2-day and 5-day old 5th instar nymphs with the help of Hamilton dispenser syringe. Control insects were treated with the same dosage of acetone (Chinzei *et al.*, 1994).

Fat body protein content of 6-day old 5th instar nymph was significantly lower than the hemolymph protein content of same age group (Fig. 1). Protein patterns of these tissues at the nymphal stage reveal the appearance of some specific bands

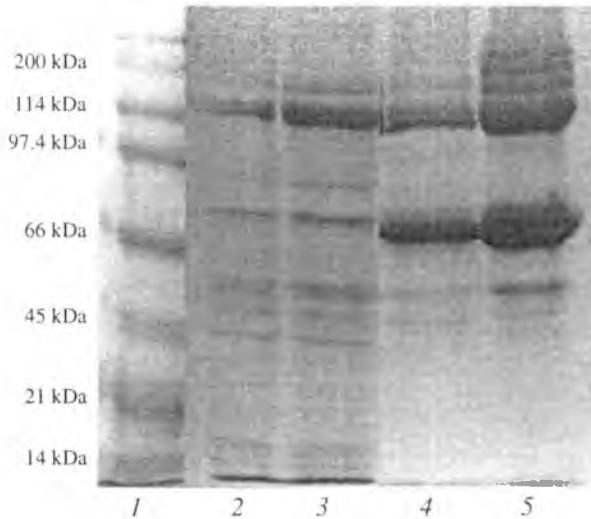


FIGURE 2. Electrophorogram showing changing protein profile in the hemolymph and fat body after Methoprene treatment. Lane 1: Marker; Lane 2: Treated fat body; Lane 3: Control fat body; Lane 4: Treated haemolymph; Lane 5: Control haemolymph.

that disappear after molting and certain bands that appear newly after adult eclosion (Fig. 1). Specific bands corresponding to MW between 21 kDa and 45 kDa present in the haemolymph of 5th instar nymph (6-day) were found to be absent in 0-day old adult. A reduction in the number of protein bands was also noticed in 0-day adults. But compared to 0-day adults, 6-day-old adult showed several more bands. Protein band of MW 114 kDa was more prominent in 6-day-old than that in 0-day-old adult. Unlike haemolymph proteins, different fat body protein bands of 6-day-old 5th instar nymphs were very feeble and weak and they did not vary significantly among themselves. However, all these protein bands are thin. The fat body of adult *Dysdercus* showed an increase in the number of bands compared to that in its 5th instar nymph. All the bands were prominent that showed equal staining intensity as well.

When methoprene ($0.5 \mu\text{g} / \mu\text{l}$) was applied on to just molted 5th instar nymphs, this emerged as supernumerary nymph (Fig. 3). Two-day-old 5th instar nymph that received $0.5 \mu\text{g}$ and $1 \mu\text{g} / \mu\text{l}$ developed into adultoids after subsequent ecdysis. When applied same dosage four days after the fifth instar nymphal molt, they prolonged the fifth instar stage for 2–3 days and more than 50% emerged into adults. The supernumerary nymphs and the adultoids retained varying degrees of nymphal characters. Wings are somewhat developed. However they do not cover the abdomen completely. Abdominal spots that are not present in normal adults were found in adultoids and supernumerary nymphs (Fig. 3).

Haemolymph and fat body were collected from methoprene-treated 5th instar nymph after 48 h and was subjected to SDS PAGE. Protein pattern of haemolymph

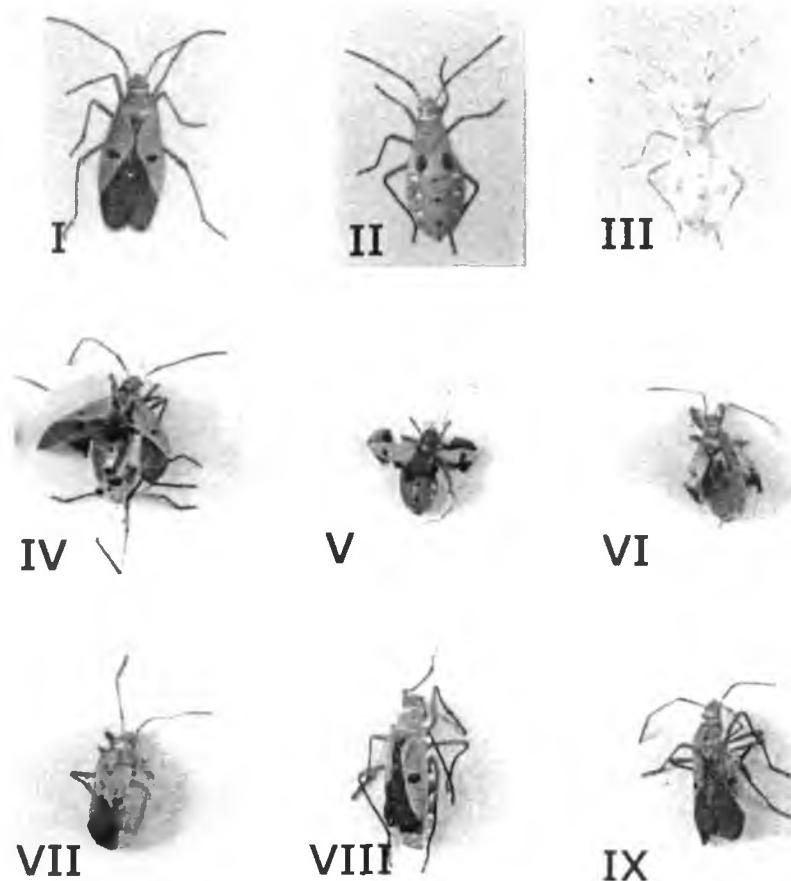


FIGURE 3. Effect of JHa on *D. cingulatus* nymphs. 1. Normal adult. 2. Normal 5th instar nymph. 3. Supernumerary nymph emerged after methoprene treatment. 4, 5, 6. Adultoids emerged after methoprene treatment showing varying deformities. 7, 8, 9. Methoprene treatment on 5-day-old 5th instar nymphs prolonged metamorphosis.

and fat body of methoprene-treated and control 5th instar nymph is as shown in Fig. 2. There is significant reduction in the intensity of almost all protein bands in the fat body of methoprene treated nymph when compared to that in the control nymph. But in the case of haemolymph protein profile, most of the proteins showed a drastic reduction in methoprene- treated nymph.

Application of JHa soon after emergence into the 5th instar nymph is found to be the most sensitive period to elicit its effect on retention of larval characters. When JHa was applied at the end of the sensitive period i.e. late fifth instar, adultoids are produced after ecdysis. Morphologically perfect extra-nymphal instars are formed when the effective concentration of the juvenoid has been maintained or surpassed during the

whole duration of the critical JH sensitive period. Different intermediate forms were found to develop when the effective juvenoid concentration has been reached for just a part of the critical period.

JHa was found to prolong the process of metamorphosis in *Trichoplusia ni* (Jones, 1985), *Xenopsylla skrjabini* (Ershova *et al.*, 1989), *Blatta Orientalis* (Short and Edwards, 1992b), *Rhodnius prolixus* (Chinzei *et al.*, 1994), *Bombyx mori* (Bharati and Miao Yungen, 2001) and *Apis mellifera* (Elekonich *et al.*, 2002). The insects which survived always completed metamorphosis by imago hatching, confirms the same results as seen in *D. cingulatus* in the present study.

Electrophoretic studies showed that there is a reduction in both fat body and haemolymph protein profile after methoprene treatment. Tiwari (1989) reported a reduction in haemolymph protein of *Diacyclos obliqua* after JHa treatment. Significant depression of total protein contents was noted in methoprene-treated pupae and adults of *Daphnia magna* when compared to its normal untreated insect (Olmstead and LeBlanc, 2001). Methoprene treatment caused a significant reduction in blood protein levels of *Locusta migratoria* (Cotton and Anstee, 1991) and also in *Heteropeza pumila* and *Mycophila speyeri* (White and Czajkowska, 2001). All these findings shows the commitment of epidermal cells to ecdysteroids that is dependent on JH activity pattern.

JH regulate the synthesis of specific larval and adult proteins in this tissue (Laufer and Borst, 1983). The mode of action of JHa lies in their stability to block the depression, transcription, or utilization of fresh genetic information (Wyatt and Davey, 1996). Juvenilizing activity of JHa is dependent on the stage of development at the time of its application. Juvenile hormone acid a catabolite of JH that induces competence of epidermal cells after ecdysteroid initiated commitment for metamorphosis (Ismail *et al.*, 1998, 2000). Studies with Juvenile hormone analogues on to 5th instar nymph suggest that metamorphosis is a sequence of events which are each dependent on the absence of JH.

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Assessment of storage losses in wheat due to stored grain insect pests in Himachal Pradesh

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ABSTRACT:

A survey conducted in Western Himachal Pradesh during 2001–2002 revealed that different storage structures used by farmers could be ranked in effectiveness as metal bin, metal drum with lid, metal drum without lid, wooden boxes and gunny bags based on percentage damage of grains and per cent loss in weight. Maximum number of damaged grains was found during rainy season and it was followed by summer and winter seasons. Similar trend was observed in per cent weight loss of wheat grains also. © 2007 Association for Advancement of Entomology

KEYWORDS: grain damage, weight loss in stored wheat

The damage caused by insects to stored food grains in tropical and temperate regions of the world is very common. Dry and cold climatic conditions have been reported to be unfavourable for majority of the stored grains pests in stores. Himachal Pradesh offers a favourable climate for storage insects. Grains are still stored by the farmers in traditional storage structures (Doharey, 1994). Agarwal *et al.* (1981) carried out a survey of the storage losses in Himachal Pradesh and since then no information is available regarding storage conditions and losses in the state. The present study was undertaken to fulfil this lacuna.

Survey for the present study was carried during 2001–2002 in two districts *viz.*, Kangra and Mandi in the state. Ninety samples were collected from each district following statistical norms. Per cent grain damage and percentage loss in weight were calculated. Per cent grain damage was calculated as per formula given by Adams and Schulten (1978). Per cent weight loss was assessed following the method of Harris and Lindblad (1978).

As evident from table 1 per cent damage to wheat grains due to insect pests varied from 2.98–13.91 and 3.06–17.90 in Kangra and Mandi district, respectively. Mean per cent damage was more during rainy season i.e. 10.53, 10.59 which was significantly

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TABLE 1. Effect of different storage structures on the percentage of wheat grain damage and weight loss due to stored grain pests

Storage structures	Kangra						Mandi					
	Percentage of grain damage			Percentage of weight loss			Percentage of grain damage			Percentage of weight loss		
	Winter	Summer	Rainy	Mean	Winter	Summer	Rainy	Mean	Winter	Summer	Rainy	Mean
Metal bin	2.98 (1.73)	5.45 (2.32)	7.35 (2.71)	5.26 (2.25)	0.93 (0.96)	1.47 (1.21)	2.36 (1.54)	1.58 (1.24)	3.06 (1.75)	5.53 (2.35)	6.68 (2.58)	5.09 (2.23)
Metal drum (with lid)	3.33 (1.82)	7.22 (2.69)	8.17 (2.86)	6.24 (2.46)	1.02 (1.01)	1.95 (1.39)	2.82 (1.68)	1.93 (1.36)	3.26 (1.80)	6.11 (2.47)	7.69 (2.73)	5.69 (2.34)
Metal drum (without lid)	3.77 (1.94)	7.44 (2.73)	9.19 (3.03)	6.80 (2.57)	1.71 (1.31)	2.59 (1.60)	3.16 (1.81)	4.40 (1.57)	3.65 (1.91)	7.29 (2.69)	9.74 (3.02)	6.89 (2.54)
Wooden box	4.11 (2.05)	8.45 (2.90)	10.99 (3.31)	7.85 (2.76)	2.14 (1.46)	3.16 (1.78)	4.40 (2.09)	3.28 (1.78)	4.11 (2.03)	9.35 (3.05)	10.94 (3.23)	8.13 (2.77)
Gunny bag	7.15 (2.67)	13.91 (3.72)	6.97 (4.12)	12.67 (3.51)	3.05 (1.75)	5.88 (2.42)	8.15 (2.86)	5.69 (2.34)	7.10 (2.66)	15.52 (3.92)	17.90 (4.32)	13.51 (3.63)
Mean	4.26 (2.04)	8.49 (2.87)	10.53 (3.21)	7.77 (1.30)	3.01 (1.68)	4.20 (2.00)	4.24 (2.03)	8.76 (2.03)	10.59 (3.18)	1.73 (1.29)	3.24 (1.75)	4.20 (2.00)
CD's	St. St. 0.016 Seasons:	0.016	St. St. 0.022 seasons:	0.017	St. St. 0.087 Seasons:	0.068	St. St. 0.017 Seasons:	0.013				

different from summer 8.49, 8.76 and winter 4.26, 4.24 % at Kangra and Mandi district, respectively. Similar was the case for per cent loss in weight which ranged from 0.93–8.15 and 0.94–8.08 at Kangra and Mandi district, respectively. Maximum loss in weight was in rainy season i.e. 4.20 at both the districts, followed by summer and winter season. Thus from the data it is evident that both the per cent damage and per cent loss in weight were more in rainy season in both the districts. Per cent damage up to 13.91 and 17.90 per cent due to insect attack has been recorded in both the districts in the present study. Bains *et al.* (1976) reported 9.02–14.48 per cent damage to stored wheat grains in Punjab whereas Srivastava *et al.* (1973) have reported up to 30.1 per cent damage to kernels due to insect attack in U.P. In the present survey per cent loss in weight in the range of 0.93–8.15 was recorded in both the districts, maximum mean percent weight loss being in rainy season (4.20%) which is in conformity with Bains *et al.* (1976), Agarwal *et al.* (1981) and Singh *et al.* (1994) in adjoining states. Girish *et al.* (1975) reported 0.06–9.7 per cent grain weight loss in stored wheat in U.P.

The storage structures differed significantly in per cent damage and per cent weight loss due to insects. Per cent mean damage was minimum in metal bin followed by metal drum with lid, metal drum without lid, wooden box and gunny bag i.e. 5.26, 6.24, 6.80, 7.85, 12.67 and 5.09, 5.69, 6.89, 8.13, 13.51 per cent at Kangra and Mandi district, respectively. Similar was the case for mean per cent weight loss which was maximum in gunny bag 5.69, 5.56 per cent and minimum in metal bin 1.58, 1.61 per cent at Kangra and Mandi district, respectively. Thus the metallic structures (metal bin and metal drum with lid) are safe for storage of food grains than the traditional structures. Present findings get support from the observations of Bains *et al.* (1976) and Dhaliwal (1977) who also recorded more damage and loss in weight in traditional local storage structures as compared to gunny and wooden boxes, which may be due to their permeability to atmospheric air and moisture which enhances insect multiplication. Metal bins and metal drums with lid are tight structures and deficient in moisture. Singh *et al.* (1994) also observed that grain losses were more pronounced in traditional local containers compared to modern storage structures. They also confirmed minimum grain weight loss in metal bins as is the case with the present findings. The dependence of storage losses on the type of storage containers used has also been confirmed beyond doubt by Prakash *et al.* (1987).

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The suitability of host plants for mass rearing of *Scirtothrips dorsalis* Hood (Thysanoptera: Thripidae)

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ABSTRACT: A mass rearing method for *Scirtothrips dorsalis* on rose flower is described. French bean pods and brinjal leaves were found unsuitable for rearing.

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KEYWORDS: *Scirtothrips dorsalis*, mass rearing

Scirtothrips dorsalis Hood (Thysanoptera: Thripidae) is a serious pest of chilli, brinjal, rose and other economically important crops in India (Ananthakrishnan, 1971; Jaganmohan *et al.*, 1980). Life stages desap tender leaves, shoots, developing flowers and fruits causing leaf curl upwards symptom (Amin, 1976), resulting in 46–90% yield loss in sweet and chilli pepper (Krishnakumar, 1995). This insect is also a key vector of tomato spotted wilt virus (TSWV) which causes bud necrosis disease (Amin *et al.*, 1981). The present study was carried out to develop a mass rearing method for *S. dorsalis*.

Scirtothrips dorsalis was obtained as follows: Terminal shoots about 10 cm long of field grown chilli plants, showing typical leaf curl symptom were cut and placed in a tall cylindrical glass jar. The mouth of the jar was closed with a muslin cloth to prevent escape of adults. The container was placed in a B.O.D incubator, at 10 °C for 10 min. to inactivate the insects. The twig was then taken out and tapped gently over a clean black platform. Most of the thrips fell down to the platform. Adults alone were collected in a clean tall glass jar (15 × 10 cm). From this sample, *S. dorsalis* were isolated based on the taxonomic characters and used for mass rearing.

Three host plants, brinjal leaves, French bean pods and rose were selected for the study. These are the natural host plants of *S. dorsalis* (Ananthakrishnan, 1971). The host plants were grown in glasshouse and parts of plants, free from any insect, including thrips, were used for the experiments.

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Rearing on French bean pods: Tender, washed and air dried pods were spread on top of a brass wire mesh placed on top of a filter paper, kept moist with wet cotton inside a plastic jar. *S. dorsalis* females were released and after two days adults were recovered and released into new rearing jars. Bean pods were kept undisturbed for emergence of larvae. Fresh bean pods were added to the jar as and when the exposed ones dried. Observations on the number of larvae, pupae and adults produced were recorded. The experiment was replicated five times with two adults per rearing jar.

Rearing on brinjal leaves: Fresh tender brinjal leaf was placed on the concave side of a watch glass flanked with a blotting paper. The edge of the leaf was trimmed, so that the leaf remained within the watch glass. This was later kept inside a circular petri plate (20 cm diameter \times 0.5 cm height) containing water. The cut end of the stalk was covered with a cotton wick that was in contact with water in the petri dish. After releasing *S. dorsalis* females on leaves, the petri plate was covered with another one of the same size. The released adults were recovered after two days and the process was repeated. Fresh leaf was kept as and when the previous one started drying. Observations on the number of larvae, pupae and adults at each stage were recorded. The experiment was replicated five times with five adults per replication.

Rearing on rose: Flowers of var. Happiness, Mulu and Gladiator were chosen. Half opened flowers which were free from any insect attack were selected. A plastic vial with lid (5 \times 3 cm) was taken and 3/4th of its capacity was filled with water. A circular hole was made in the centre of the lid, through which the flower stalk was inserted. A petri plate (20 width \times 0.5 cm height) was taken and a watch glass was kept centrally with the concave surface facing upwards. The plate was filled with water up to half its capacity. The plastic vial with the flower was placed on top of the watch glass. The flower was covered with a glass chimney (10 \times 15 cm width/height). After releasing *S. dorsalis* the other end was covered with a tissue paper held tight by rubber band.

To record the progeny production, the exposed flower was taken out at weekly intervals and tapped gently on to a watch glass (15 cm diameter) that was kept inside a petri plate (25 \times 0.5 w/h) filled with water. Majority of thrips fall to the watch glass. Tapping was continued for two or three times till no more thrips fell down. The flower was later replaced into the rearing chamber and observation was repeated after a week by following the same procedure.

The number of larvae and pupae falling to the watch glass was recorded. Larvae were released into fresh rearing chambers for further development, while pupae were kept on a rose petal inside a Petri-plate (10 cm diam.) having a moist filter paper, folded into two or three layers, for adult emergence. The experiment was carried out with two females per replication and each treatment was replicated six times.

Among the three host plants tried, french bean pod and brinjal leaves were found to be unsuitable for multiplication of *S. dorsalis*. In french bean pods, though released females were observed to remain alive up to a week, no larvae could be recorded indicating absence of oviposition. No larvae were recorded in brinjal leaf also. The insects multiplied on rose flower. Among the rode varieties, Gladiator and Mullu were

TABLE 1. Progeny production and per cent survival of *S. dorsalis* at different generations

Generations	No of larvae recorded	No. of pupae recorded	No of adults recorded
1st	98.40	87.60	73.40
2nd	83.00	73.80	67.20
3rd	85.40	87.20	61.40
5th	85.60	73.20	61.00
10th	81.20	73.90	59.00
CV	10.69	14.76	22.17
CD @ 0.05%	13.02	15.66	19.14

found to be equally suitable. Happiness flowers wilted within two or three days and were not suitable for mass rearing.

Preliminary studies on variety Gladiator, showed that releasing two females per flower yielded more progeny (60 nymphs/female) as compared to releasing five (29 nymphs/flower) or ten females per flower (16 nymphs/female). Hence further studies were carried out by releasing two females per flower.

Progeny production in different generations are presented in table 1. The data showed no deterioration in progeny production up to 10th generation.

Observations on rose flowers showed that larval stages were more in inner petals than in outer petals. Pre-pupae and pupae occurred on the undersurface of sepals, on curled tips of petals and on the base of petals. Normally pre-pupae and pupae were found aggregated together while larval stages were dispersed on the flower.

As already noted among the host plants studies, rose var. Gladiator was the best for rearing of *S. dorsalis*. Regular changing of flower was not needed as a single flower could support all stages of *S. dorsalis* for more than 10 days, without wilting. As and when the exposed flower starts drying fresh flower was kept on top of the earlier one. Thrips migrated within 1–2 h, to the new flower. After 24 h the old flower was removed and culture was continued on the fresh one.

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Ant fauna associated with whiteflies in some locales of Eastern and Western Ghats in two Southern states of India

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ABSTRACT: A list of whiteflies attracting ant species on wide range of flora in some localities of Eastern and Western Ghats of Tamil Nadu and Kerala is provided. The ecotypic diversity of ant tending whiteflies such as *Aleuromarginatus* sp., *Aleuroclava* sp., *Sphericaleyrodes indica*, and *Rhachisphora* sp. on the host plants was influenced by species of tending ants. The whitefly, *Aleuroclava* sp. inviting ant species were totally different in the same host plant of distant localities whereas the whitefly *Aleuromarginatus* sp. attracting ant species in the nearby localities were similar. The whitefly *Rhachisphora* sp. attracted diverse ant species from different localities.

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KEYWORDS: mutualistic interaction, ant tending whitefly, Western Ghats

Understanding the interactions between species or populations is a prerequisite for predicting ecological phenomena at all levels of biological organization. For this respect, mutualistic interactions have generally received less attention as compared to competition and predation in the concept of species existence and survival among living organisms. In most mutualisms, one partner performs some service that benefits its associate, and in return receives some pay-off or reward. The local biotic environment, that is, the identity and abundance of other species with which the mutualists interact, also strongly influences outcomes. Costs and benefits of anti-herbivore mutualisms are influenced by the herbivores' host plant (Stadler *et al.*, 2001).

Hemipterans infesting a wide range of host plants exude honeydew, which attract a number of ants belonging to the Subfamilies Dolichoderinae, Formicinae, Myrmicinae and Pseudomyrmicinae (Veeresh and Ali, 1990; Rastogi *et al.*, 1997). With some aleyrodids species viz., *Rhachisphora*, *Sphericaleyrodes*, *Aleuromarginatus*, *Aleuroclava*, *Aleurolobus* and *Dialeurodes*, ants are found frequently associated for honeydew and wax (Jesudasan and David, 1995). Whiteflies benefit from ant tending because the presence of ants deter insect predators from their host plants, increase their feeding

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TABLE 1. Whiteflies attracting ant species in some localities of Eastern and Western Ghats

Location/Host plants	Whitefly	Ants and their subfamilies
Aliyar/ <i>Grewia</i> sp.	<i>Aleuromarginatus</i> sp.	Formicinae <i>Anoplolepis gracilipes</i> Jerdon <i>Camponotus compressus</i> Fabr. <i>Camponotus rufoglaucus</i> Jerdon Myrmicinae <i>Crematogaster</i> sp. Ponerinae <i>Pachycondyla</i> sp. Pseudomyrmicinae <i>Tetraponera nigra</i> Jerd
Top Slip/ <i>Grewia</i> sp	<i>Aleuromarginatus</i> sp.	Formicinae <i>Anoplolepis gracilipes</i> Jerdon <i>Camponotus compressus</i> Fabr. <i>Camponotus rufoglaucus</i> Jerdon Myrmicinae <i>Crematogaster</i> sp. Ponerinae <i>Pachycondyla</i> sp.
Mannuthu/ <i>Cassia</i> sp.	<i>Aleuroclava</i> sp.	Dolichoderinae <i>Tapinoma melanocephalum</i> Fabr. Formicinae <i>Camponotus compressus</i> Fabr. Myrmicinae <i>Crematogaster rothneyi</i> Mayr <i>Monomorium indicum</i> Fornel <i>M. latinode</i> Mayr Pseudomyrmicinae <i>Tetraponera rufonigra</i> Jerdon
Edapalyam/ <i>Cassia</i> sp.	<i>Aleuroclava</i> sp.	Formicinae <i>Paratrechina longicornis</i> Latr. <i>Monomorium</i> sp.
Mekkarai/ <i>Ochlandra travancorica</i>	<i>Sphericaleyrodes indica</i>	Dolichoderinae <i>Techomyrmex albiceps</i> Smith Formicinae <i>Acantholepis</i> sp. <i>Anoplolepis gracilipes</i> Jerdon <i>Anoplolepis longipes</i> Fabr. <i>Paratrechina</i> sp. <i>Polyrhachis illudata</i> Walker <i>P. rastallata</i> Latr <i>P. exercita</i> Walker Myrmicinae <i>Catulacus taprobannea</i> Smith

TABLE 1. (Contd...)

Location/Host plants	Whitefly	Ants and their subfamilies
Aryankavu/ <i>Ochlandra travancorica</i>	<i>Sphericaleyrodes indica</i>	Formicinae <i>Anoplolepis gracilipes</i> Jerdon <i>Anoplolepis longipes</i> Fabr. <i>Paratrechina longicornis</i> Latr. <i>Polyrhachis</i> sp. <i>Monomorium</i> sp.
Pamalakavlu/ <i>Ochlandra travancorica</i>	<i>Sphericaleyrodes indica</i>	Dolichoderinae <i>Techomyrmex albiceps</i> Smith Formicinae <i>Anoplolepis longipes</i> Jerdon <i>Polyrhachis rastallata</i> Latr. <i>Oecophylla smaragdina</i> Fabr. Myrmicinae <i>Myrmicaria brunnea</i> Saunders Pseudomyrmicinae <i>Tetraponera aitkeni</i> Forel
Damdam Parai/ <i>Ixora pavetta</i>	<i>Rhachisphora</i> sp.	Dolichoderinae <i>Techomyrmex</i> sp. <i>Tapinoma melanocephalum</i> Fabr. Formicinae <i>Camponotus compressus</i> Fabr. Myrmicinae <i>Crematogaster</i> sp. <i>Myrmicaria brunnea</i> Saunders
Megamalai <i>I. pavetta</i> , <i>Memecylon umbellatum</i> , <i>Syzygium cumini</i>	<i>Rhachisphora</i> sp.	Dolichoderinae <i>Tapinoma melanocephalum</i> Fabr. Formicinae <i>Camponotus compressus</i> Fabr. Myrmicinae <i>Crematogaster rothneyi</i> Mayr. <i>Monomorium indicum</i> Forel <i>M. latinode</i> Mayr Pseudomyrmicinae <i>Tetraponera rufonigra</i> Jerdon

rates, and prevent the buildup of fungus from collected honeydew (Delabie, 2001). However, the nature of interaction can exhibit considerable spatial variation (Cushman *et al.*, 1998), and it is therefore important to observe this interaction across such special scale. Hence, a study was undertaken to collect and identify the whiteflies and ants from some host plants occurring in different localities of Eastern Ghats and Western Ghats of Tamil Nadu and Kerala, India.

TABLE 1. (Contd...)

Location/Host plants	Whitefly	Ants and their subfamilies
Tambaram <i>I. pavetta,</i> <i>M. umbellatum,</i> <i>Syzygium cumini</i>	<i>Rhachisphora</i> sp.	Dolichoderinae <i>Tapinoma melanocephalum</i> Fabr. Formicinae <i>Camponotus paria</i> Emery <i>Camponotus rufoglaucus</i> Jerdon <i>Camponotus</i> sp. nr. taylori <i>Oecophylla smaragdina</i> Fabr. <i>Paratrechina longicornis</i> (Latr.) <i>Polyrhachis exercita</i> (Walker) <i>P. scissa</i> Roger Myrmicinae <i>Crematogaster dalyi</i> Forel <i>Meranoplus bicolor</i> Guer <i>Monomorium indicus</i> Forel <i>Myrmicaria brunnea</i> Saunders. <i>Pheidole</i> sp. Pseudomyrmicinae <i>Tetraponera aitkeni</i> (Forel) <i>Tetraponera nigra</i> Jerdon <i>Tetraponera rufonigra</i> (Jerdon)

Collection of leaves bearing whitefly tending ants were made from Tambaram (Selaiyur range), Damdam Parai (Palani Hills), Aryankavu and Edapalayam (Thenmalai range), Mekkarai (Tenkasi range), Mannuthu (Megamalai range), Aliyar (Valparai range), Top slip (Anaimalai Hills), Pamalakavlu (Adimali range), on the host plants viz., *Ixora pavetta*, *Memecylon umbellatum* (Burman), *Syzygium jambolanum* (Lam.), *S. cumini*, *Grewia* sp., *Cassia* sp. and the reed, *Ochlandra travancorica* (Bedd.).

The ecotypic diversity of ant tending whiteflies such as *Aleuromarginatus* sp., *Aleuroclava* sp., *Sphericaleyrodes ochlandrae*, and *Rhachisphora* sp. on the host plants was influenced by species of tending ants (Table 1). The ant, *Anoplolepis longipes* was commonly tending the whitefly host plants on all the three localities whereas *Catulacus taprobannea*, *Acantholepis* were only found in Mekkarai. The ant *Techomyrmex albiceps* was tending the whitefly, *Sphericaleyrodes indica* in Mekkarai and Pamalakavlu localities whereas *Oecophylla smaragdina*, *Myrmicaria brunnea* and *Tetraponera aitkeni* were only seen in Pamalakavlu local whitefly species. The ants such as *Polyrhachis* sp. and *Paratrechina* sp. were tending the whitefly of Mekkarai and Aryankavu where as *Monomorium* sp. was only noticed in Aryankavu site host plant. Intraspecific variation in host-plant quality as influenced by environmental conditions might affect herbivore-ant interactions (Pierce *et al.*, 1991).

The ants attending to the whitefly, *Aleuroclava* sp. were totally different on same host plants from distant localities such as Mannuthu (Magamalai) and Edapalyam (Thenmalai) whereas the whitefly *Aleuromarginatus* sp. attracting ant species in the

nearby localities viz., Aliyar (Valparai), Top slip (Anaimalai) of the host plant, *Grewia* sp., were almost similar (Table 1). Cushman *et al.* (1998) stated that some members of homopteran species were significantly more to attract ants than those of others.

Diverse ants have been noted to be associated with the whitefly, *Rhachispora* sp. from different localities (Table 1). The *Tapinoma* sp., *Crematogaster* sp. and *Camponotus* sp. were common in occurrence in all the localities (Damdam Parai, Mannuthu and Tambaram). Similarly, Cushman and Addicott (1991) reported that homopteran attracting ants were often varied in their assemblages and taxonomic diversity across large geographic spatial scale.

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Histochemical study of yolk granules of ripe oocytes of *Oxya hyla hyla* (Orthoptera: Acrididae)

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ABSTRACT: Isolated yolk granules of vitellogenic oocyte of *Oxya hyla hyla* under light microscope revealed the presence of deeply stained proteinaceous rim region and lightly stained core region. The same granules were also positive for carbohydrate and lipid. Those yolk granules initially deposited in smaller size gradually showed increase in size due to aggregation of different components.

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KEYWORDS: *Oxya hyla hyla*, ripe oocyte, yolk granules, histochemistry

Vitellogenesis is one of the key events occurring in growing oocytes of both vertebrates and invertebrates. This process is usually under control of several hormones (Courceroles and Kondo, 1980). Although a good number of works have been published on purification and characterization of yolk proteins (vitellin, lipovitellin) (Harnish and White, 1982) including that of Locust (Chen *et al.*, 1978) sufficient attention has not been paid to study the yolk granules taken from the ripe oocyte in its original condition under microscope. Raikhel (1984) using high-resolution immuno- and cytochemical study had shown that in mosquito (*Aedes aegypti*) oocytes the yolk granules are initially of low electron density but gradually grow larger in size and of higher electron density. *Oxya hyla hyla* (grasshopper) is an important orthopteran paddy pest of Tripura. Depending on structure and physiology, the ovariole of *Oxya* is panoistic in nature and is without any nurse cells. Ghosh *et al.* (1997) by histological studies had shown that in growing oocytes of *Gesonula punctifrons*, the small sized yolk droplets initially deposited near the follicular epithelium slowly grew larger in size. Therefore an effort was made to study the isolated yolk granules under light microscope using various cytochemical methods.

Ripe terminal oocytes were collected from the mature females of *Oxya hyla hyla* collected from paddy fields around Agartala city. For histochemical studies, yolk granules were obtained by rupturing the fully ripe oocytes over a slide and

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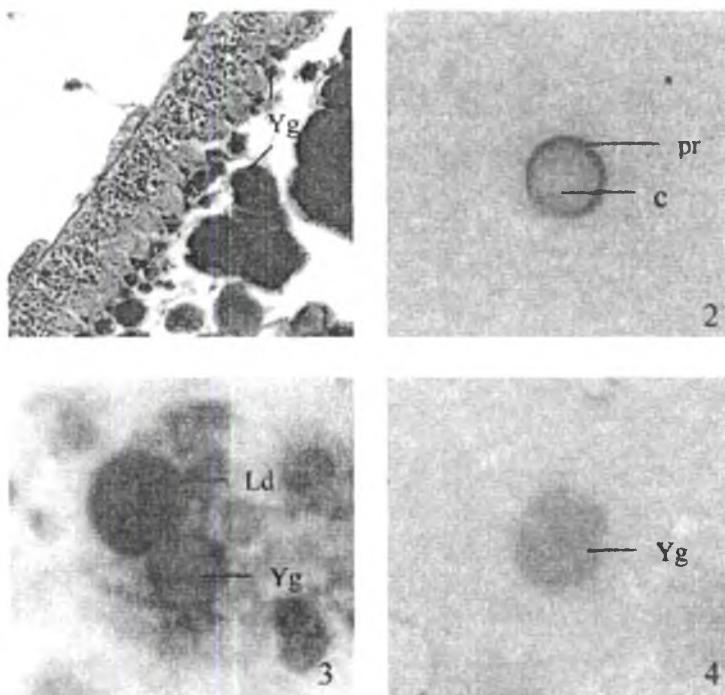


FIGURE 1. Photomicrograph of sectioned vitellogenic oocyte of *Oxya hyla hyla* stained with Hg-Bromphenol Blue showing the yolk granules (Yg) of different sizes. 350X.

FIGURE 2. Photomicrograph of the separated yolk granules (Yg) stained with Hg-Bromphenol Blue showing differential stainability of the peripheral rim (pr) and lightly staining core (c). 330X.

FIGURE 3. Photomicrograph of the deeply stained Sudan Black B positive granules (Ld) depicting the lipoidal nature of the yolk granules. 330X.

FIGURE 4. Photomicrograph showing the PAS positive nature of the overall granule depicting presence of carbohydrate substance in the yolk granules. 390X.

spreading its contents homogeneously over it. Then those yolk contents which were seen to contain spheres in unstained condition under microscope were fixed in Ca-formol solution and air dried for 24 h or more. Those slides were used for further histochemical studies. To examine the presence of proteins in the yolk granules lipids were extracted from this slide by methanol-chloroform (1:1) extraction method. Protein moiety in the granules was stained by Hg-Bromphenol blue method (Pearse, 1975). To stain lipid granules, Sudan black B was used. In this staining method, three sets of Ca-formol fixed dried slides containing the yolk granules (one set treated with methanol-chloroform extraction method, one set treated with cold acetone extractin method and one set without any extraction) were first immersed in 2.5% aqueous Bromine solution for 30 min and then washed in distilled water for 30 min. Then

the slides were immersed in 70% alcohol for 30 min and stained in saturated Sudan Black B solution in 70% alcohol. Excess stain was removed by washing in distilled water (Pearse, 1975). To stain mucoid substances in the yolk granules Schiff's reagent was prepared and staining was done after oxidation by Periodic acid solution (Pearse, 1975).

The yolk granules when stained with Hg-Bromphenol blue showed intensely Hg-Bromphenol blue positive reaction. The diameter of those granules were about $32.8 \pm 9.3 \mu\text{m}$. It was also observed that those Hg-Bromphenol bule positive granules showed differential staining reaction in its various zones (Fig. 1, 2). The inner core region of those granules became faintly stained while the outer rim became deep blue. The diameter of the core region was about $22 \pm 5 \mu\text{m}$ and the outer rim was about $5.5 \pm 2 \mu\text{m}$ thick. This result indicated that the granules were composed of different types of protein components. The basic protein originally becomes more insensly stained in comparison to acidic or neutral proteins (Mazia *et al.*, 1953). So it was apparent that the outer rim of those protein granules were made up of proteins with more basicity and in the inner core region basicity was low. Again when those yolk granules were stained with Sudan Black B the granules treated with methanol-chloroform and cold acetone extraction methods showed negative reaction but the set without any extraction showed strong positive reaction and became brownish black in colour (Fig. 3). This result suggested the presence of lipid or lipoprotein in the yolk granules. Further when stained by PAS reaction after oxidation with Periodic acid those granules of almost same diameter showed PAS positive reaction and became faint magenta in colour (Fig. 4). Such positive reaction suggested that those yolk granules also contained carbohydrate moiety.

All the results taken together strongly suggested that the yolk granules or the droplets which were present in the oocyte had a complex nature and constituted of protein, carbohydrate and lipid moieties. In the sectioned condition (Fig. 1) these granules were found in different sizes and in isolated condition also, that size variation was apparent. The variation of the size of the yolk granules strongly suggested that there was a morphogenetic pathway in the development of the large size yolk granules from the smaller. That may be due to aggregation of different components or other phenomena may be involved. Raikhel (1984) using high resolution immuno and cytochemical electron microscopy have shown, a similar pathway in mosquito oocyte. He has also indicated that in the yolk granules some crystalline and some noncrystaline protein part was present. In the present situation, the presence of proteinaceous core and the rim suggested the same phenomena.

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